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# Bulletin

*of the*

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# Plantae Krukovianae

H. A. Gleason and A. C. Smith

(*concluded from page 365*)

## Sterculiaceae

**Sterculia megalocarpa** A. C. Smith, sp. nov. Arbor ad 25 m. alta, trunco circiter 1 m. diametro; ramulis crassis cinereis striatis, juventute ferrugineo-pilosis mox glabris; stipulis subcoriaceis glabrescentibus lanceolatis 10–17 mm. longis; foliis ad apices ramulorum congestis; petiolis semiteretibus striatis 2.5–6 (rare ad 8) cm. longis, basi apiceque incrassatis, pilis adpressis ferrugineis strigosis; foliis integris coriaceis opacis oblongis vel ovato-oblongis, 12–21 cm. longis, 6–11 cm. latis, basi rotundatis saepe subcordatis, apice cuspidatis vel breviter acuminatis, margine integris undulatis, supra glabris, subtus densissime et arcte ferrugineo-tomentosis (pilis stellatis 4-ramosis 0.2–0.3 mm. latis), pinnatinerviis (e basi 5- vel 7-nerviis), nervis lateralibus 10–12 in quoque latere rectis adscendentibus prope margines anastomosantibus, cum costa supra elevatis subtus prominentibus, venulis reticulatis supra planis subtus elevatis; inflorescentiis desideratis; carpidiis vetustis maximis oblongo-ovoideis falcatis, 15–20 cm. longis, 9–12 cm. latis, juventute arcte ferrugineo-tomentosis, maturitate glabris, breviter stipitatis, pericarpio fibroso sutura 2.5–4.5 cm. crasso (Pl. 21, f. 1).

Type, *Krukoff 1675*, collected Dec. 27, 1931, on "terra firma" near the source of the Jatuarana River, Machado River region, State of Matto Grosso. The plant is locally known as "Achicha." It is a species distinguished from other South American *Sterculiae* by its extraordinarily large fruit, which is comparable only to the fruit of *S. Chicha* St. Hil., which, however, has lobed leaves. *S. megalocarpa* is allied to *S. pruriens* (Aubl.) Schum., *S. pilosa* Ducke, and *S. Tessmannii* Mildbr. From *S. pruriens* the present species differs by its pubescent leaves, from *S. pilosa* by its short-acuminate leaves. *S. Tessmannii* is said to have longer petioles, longer-acuminate leaves, and fewer lateral nerves than our species. The fruits of the first two of these species are known to be much smaller than ours; of *S. Tessmannii* the fruit is not yet known.

## Guttiferae

**Calophyllum angulare** A. C. Smith, sp. nov. Arbor ad 30 m. alta, trunco 1 m. diametro; ramulis crassis fuscis, juventute quadrangularibus et fusco- vel cinereo-puberulis, internodiis 1–2 cm. longis; petiolis saepe nigrescentibus rugosis leviter canaliculatis glabris 13–20 mm. longis; laminis rigide coriaceis

[The Bulletin for May (60: 323–378) was issued May 1, 1933.]

glabris oblongis vel ovato-oblongis, 8–11 cm. longis, 3.5–5.5 cm. latis, basi acutis vel cuneatis, apice breviter acuminatis, margine integris incrassatis, costa supra saepe impressa subtus prominentissima et striata, nervis lateralibus numerosissimis densissimis parallelis; inflorescentiis axillaribus racemosis vel e cymis brevibus compositis, 2–5 cm. longis, 6–15-floris, ubique dense ferrugineo-puberulis; bracteolis ovatis 4–6 mm. longis mox deciduis; pedicellis 2–5 mm. longis; sepalis 4 intus glabris ovatis, 5–6 mm. longis, 3 mm. latis; petalis plerumque nullis interdum 2, sepalis similibus sed glabris et membranaceis; staminibus numerosis, filamentis gracilibus circiter 2.5 mm. longis, antheris 1 mm. longis; ovario glabro ovoideo sub anthesi 1.5 mm. longo, stylo quam ovario breviore, stigmate truncato.

Type, *Krukoff 1442*, collected Nov. 25, 1931, on varzea land near rivershore, near Tabajara, upper Machado River region, State of Matto Grosso. The collector notes that the local name is "Jacareuba." The flowers are white, and the inner bark yields a yellow latex. "Jacareuba" is also applied to the other species of the genus, notably *C. brasiliense* Camb., from which the present species differs in aspect of inflorescence. It is more closely allied to *C. pachyphyllum* Pl. & Tr., with which it has in common a ferruginous-puberulous tomentum on the inflorescence, a character which distinguishes these two species from others in South America. The present species differs from *C. pachyphyllum* by the slightly smaller leaves which are short-acuminate rather than obtuse or emarginate at apex, the longer inflorescence with more numerous flowers, and the lack of petals. *C. pachyphyllum*, which is represented by *Krukoff 1495* from the same region as the new species, usually has 4 petals; *C. angulare* is usually apetalous, rarely with 2 petals.

## CARYOCARACEAE

**Caryocar dentatum** Gleason, sp. nov. Arbor 20 m. alta; ramuli subteretes crassi, 4 dm. infra apicem 5 mm. diametro fistulosi, tenuiter brunneo-pubescentes, internodiis 35–45 mm. longis; petioli inferiores 65 mm. superiores 25 mm. longi puberuli, stipellis 1–3 subulatis curvatis 3 mm. longis ornati; petioluli puberuli 4–7 mm. longi inter se subaequales; laminae firmulae anguste oblongae vel ellipticae, in quoque folio fere aequales, majores ad 120 mm. longae 45 mm. latae, juniores minores, basi obtusae vel late cuneatae, apice breviter acuminatae, margine grosse dentatae, dentibus triangularibus obtusis sursum vergentibus saepissime 5–8 mm. inter se distantibus et 2–3 mm. altis, venis venulisque supra paullum impressis subtus elevatis, tenuissime et pulcherrime reticulatis, secundariis in $\frac{3}{4}$ longitudinis subrectis prope marginem arcuato-conjunctis, pagina superiore ad venam mediam minutissime puberula ceterum glabra, inferiore puberula ad venas et venulas majores; pedunculus crassus puberulus 35 mm. longus; racemus 4 cm. longus circa 20-florus,

rhachide pedicellis et calycibus velutino-tomentellis; pedicelli adscendentes ad 15 mm. longi, prophyllis mox delapsis prope basin sitis; calyx late hemisphaericus in alabastro maturo 7 mm. latus, basi in pedicellum crassum 2 mm. longum decurrens, lobis 5 late triangulari-ovatis 2 mm. longis 3 mm. latis obtusis; petala oblongo-elliptica 25 mm. longa 9 mm. lata obtusa; stamina numerosa (usque 100?), filamentis 6 cm. longis carmesinis; fructus ignotus; nuces subglobosae 6 cm. longae 5 cm. latae densissime spinosae aculeis 15 mm. longis.

Type, *Krukoff 1294*, collected Nov. 7, 1931, on "terra firma" near Calama, Madeira River region, State of Amazonas. While a few other species of *Caryocar* have similarly narrow leaves, in none of them are they so deeply dentate. This feature together with the tomentose pedicels and calyx, the pubescent petiole, and the nearly basal prophylls serves to separate *C. dentatum* from all other described species of the genus.

### Lecythidaceae

**Lecythis jarana** (Huber) A. C. Smith, comb. nov. *Chytroma jarana* Huber, Bol. Mus. Goeldi **6**: 209. 1910, nomen. *Holopyxidium jarana* Ducke, Arch. Jard. Bot. Rio **4**: 152. 1925. *Eschweilera jarana* Ducke, Arch. Jard. Bot. Rio **5**: 177. 1930.

**Lecythis jarana** var. **latifolia** (Ducke) A. C. Smith, comb. nov. *Eschweilera jarana* var. *latifolia* Ducke, Arch. Jard. Bot. Rio **5**: 178. 1930.

Pará: upper Cupary River, plateau between Xingu and Tapajos Rivers, *Krukoff 1213*. Mr. Krukoff notes his plant as a tree 30 meters high and with a trunk slightly more than 1 meter in diameter, with the local name of "Jarana." Study of the flowers makes necessary the above new combination. The vertex of the ovary is more or less flat, bearing in the center a style about 2 mm. long; the 4-celled ovary has a central column, upon which the several ovules are borne on distinct funicles. These characters are those of *Lecythis* rather than of *Eschweilera* (sect. *Chytroma*). In addition, the general aspect of the plant, of which the comparatively thin leaves do not resemble those of *Eschweilera*, is that of *Lecythis*. Fruits such as those described and figured by Ducke[1] are certainly of *Lecythis* rather than *Eschweilera*. These fruits were probably taken from *L. jarana* var. *latifolia* rather than from *Eschweilera retusa* (Berg) Nied., as implied by Ducke in his second publication[2] discussing this alliance.

**Eschweilera** (*Eueschweilera*) **polyantha** A. C. Smith, sp. nov. Arbor 20 m. alta, trunco ad 1 m. diametro; ramulis cinereis vel fuscis teretibus parce lenticellatis; petiolis rugosis leviter canaliculatis 8–10 mm. longis; laminis

[1] Arch. Jard. Bot. Rio **4**: 152. *pl. 15* (as *Holopyxidium retusum*). 1925.
[2] Arch. Jard. Bot. Rio **5**: 178. 1930.

coriaceis olivaceis glabris oblongis, 12–17 cm. longis, 4–7 cm. latis, basi obtusis vel cuneatis, apice caudato-acuminatis, margine obsolete serrulatis, pinnatinerviis, costa valida, nervis lateralibus primariis 8–10 in quoque latere arcuato-adscendentibus prope margines anastomosantibus, utrinque elevatis, venulis copiose reticulatis supra leviter elevatis subtus prominulis; paniculis terminalibus et axillaribus (1–4 in axillis) ramosissimis 2–8 cm. longis multifloris; rhachidibus et ramulis subteretibus dense fusco-puberulis; pedicellis teretibus 2.5–3.5 mm. longis velut rhachidibus puberulis; sepalis carnosis minute puberulis oblongis, circiter 3 mm. longis et 1.5 mm. latis, apice obtusis, margine parce glandulosis; petalis submembranaceis glabris luteis ovatis, 7–9 mm. longis, 5–6 mm. latis, apice rotundatis; androphoro explanato 8–10 mm. longo, ligula 2 mm. lata basi dilatata, galea subsphaerica 4–5 mm. diametro, fimbriata et subtus densissime echinata, appendiculis anantheris imbricatis linearibus 2.5–3.5 mm. longis obtecta; staminibus circa annulum et in ligula prope basin numerosis (60–120), filamentis gracilibus 0.5–0.8 mm. longis apice angustis, antheris subglobosis circiter 0.4 mm. diametro; ovario semisupero, vertice depresso-conico rugoso umbonato, loculis 2, ovulis in quoque loculo 4 vel 5 erectis e basi sessilibus.

Type, *Krukoff 1318*, collected Nov. 12, 1931, on "terra firma" near Tabajara, upper Machado River region, State of Matto Grosso. The local name is "Kipeiro," a name which is apparently not used for other Lecythidaceae in the region. It is allied to several small-flowered species from Guiana, *E. parviflora* (Aubl.) Miers, *E. micrantha* (Berg) Miers, and *E. Sagotiana* Miers. From these species *E. polyantha* differs by its copiously-branched many-flowered inflorescences, and by the less conspicuous venation on the lower surface of leaves.

**Eschweilera** (*Jugastrum*) **truncata** A. C. Smith, sp. nov. Arbor glaberrima 15–20 m. alta; ramis ramulisque cinereis teretibus striatis lenticellatis; petiolis crassis rugosis canaliculatis 12–20 mm. longis; laminis coriaceis olivaceis opacis oblongis, 15–40 cm. longis, 5–12 cm. latis, basi obtusis vel cuneatis, apice acuminatis (acumine ad 2 cm. longo), margine leviter recurvatis et integris vel obsolete serrulatis, subtus parce nigro-punctatis, pinnatinerviis, costa crassa utrinque prominentissima, nervis lateralibus primariis 10–18 in quoque latere prope margines anastomosantibus, supra elevatis subtus prominentibus, venulis copiosissime reticulatis utrinque elevatis; paniculis plerumque terminalibus ramosis 5–16 cm. longis; rhachidibus fuscis angulatis rugosis lenticellatis, geniculis incrassatis; pedicellis crassis 8–10 mm. longis; sepalis subaequalibus carnosis laevibus ovato-oblongis, 5–7 mm. longis, 4.5–5.5 mm. latis, apice rotundatis, margine membranaceis; petalis tenuiter carnosis ovatis, 8–17 mm. longis, 6–13 mm. latis, apice rotundatis; androphoro explanato 20–25 mm. longo, ligula 5–8 mm. lata, galea carnosa 7–10 mm. diametro, margine fimbriata et subtus dense echinata, appendiculis anantheris imbricatis

oblongo-linearibus 2–4 mm. longis obtecta, staminibus circa annulum numerosissimis, filamentis carnosis circiter 1.5 mm. longis apice angustis, antheris oblongo-ovoideis 0.5 mm. longis; ovario subsupero, vertice conico laevi, stylo brevi obtuso, loculis 2, ovulis in quoque loculo 6–10 angulatis erectis e basi sessilibus; pyxidio subgloboso basi truncato apice turbinato, prope basin zona calycari e sepalis coriaceis acutis ad 7 mm. longis nexis cincto; vitta interzonali suberecta 6–10 mm. longa; zona superiore circulari integra; operculo convexo 15–25 mm. longo, obtuse umbonato, intus septo persistente signato; pericarpio fusco granuloso-rugoso obsolete biloculari; seminibus plerumque 3 angulatis, apice truncatis, maturitate 15–18 mm. longis (Pl. 21, f. 2).

Type, *Krukoff 1531*, collected Dec. 5, 1931, on "terra firma" near Angustura, near the source of the Jatuarana River, Machado River region, State of Matto Grosso. Another collection from the same locality is *Krukoff 1550*. *Krukoff 1382*, collected near Tabajara in the same region, also represents the species. The two latter specimens bear the mature fruit above described. The flowers are noted as white, the local name as "Matámatá." Possibly this is the same species described by Miers as *Jugastrum subcinctum*, of which only the fruit is known. However, in the present species (although at anthesis numerous ovules are observed) apparently only 3 or 4 seeds mature, while the seeds of *Jugastrum subcinctum* are said to be many. The pyxidium here described is very similar to that described and figured by Miers. The truncate-based pyxidia and the nearly basal calycalis zones of *E. truncata* and *Jugastrum subcinctum* distinguish them from other species of the section.

**Couratari macrosperma** A. C. Smith, sp. nov. Arbor excelsa glabra ad 35 m. alta, trunco 1–1.5 m. diametro; ramis ramulisque teretibus striatis cinereis vel juventute fuscis; foliis prope apices ramulorum brevium alternatis, petiolis crassis nigrescentibus rugosis profunde canaliculatis 7–9 mm. longis, laminis coriaceis oblongis vel obovato-oblongis, 11–20 cm. longis, 5–9 cm. latis, basi acutis, apice rotundatis et breviter acuminatis vel cuspidatis, margine leviter crenatis, inter costam et margines sulculis 2–4 angustis curvis longitudinalibus signatis, pinnatinerviis, costa prominentissima, nervis lateralibus primariis 17–22 in quoque latere rectis patulis prope margines anastomosantibus; inflorescentiis desideratis; pyxidio cylindrico-conico, 15–23 cm. longo, zona calycari 7.5–10.5 cm. diametro 6-gona leviter elevata interrupta, vitta interzonali suberecta 1–2 cm. longa, operculo laevi 7–9 cm. diametro centro leviter depresso, columella crassa subtriquetra; seminibus compressis oblongo-lanceolatis, circiter 13 cm. longis et 2.5 cm. latis, scuto embryonifero centrali circiter 6 cm. longo et 1.2 cm. lato, ala membranacea cincto (Pl. 22, f. 1, 2).

Type, *Krukoff 1513*, collected Dec. 3, 1931, on "terra firma" near Tabajara, upper Machado River region, State of Matto Grosso. The collector

notes that the plant is locally known as "Tauary." It is a species related to *C. glabra* Camb., of southern Brazil, from which it differs by its larger leaves and larger pyxidium, by its smooth rather than sulcate operculum, and by its seeds, which are three times as long and differently shaped. It may also be related to *C. Tauari* Berg, of which neither fruit nor flowers are known, but which is said to have long-acuminate leaves with fewer nerves than the present species.

**Couratari Krukovii** A. C. Smith, sp. nov. *C. macrospermae* affinis, habitu et foliis simillimis, sed pyxidio et seminibus differt; pyxidio conico-ellipsoideo, 11–14 cm. longo, ad medium 5.5–7 cm. diametro, zona calycari 4–5.5 cm. diametro 6-gona continua, sepalis coriaceis saepe persistentibus, vitta interzonali convexa angustissima 3–5 mm. longa, operculo 3–3.5 cm. diametro centro depresso, columella crassa triquetra 8–10 cm. longa; seminibus compressis obovato-oblongis, 8–9 cm. longis, 2.5–3 cm. latis, scuto embryonifero centrali 4.5–5 cm. longo et 1.2–1.5 cm. lato, ala membranacea cincto (Pl. 22, f. 3, 4).

Type, *Krukoff 1653*, collected Dec. 22, 1931, on "terra firma" near the source of the Jatuarana River, Machado River region, State of Matto Grosso. The local name is noted as "Tavary," which is probably interchangeable with "Tauary." In foliage there is practically no difference between this species and the preceding, except that the leaves of *C. macrosperma* average slightly larger. The two species seem very distinct from others of the genus by the conspicuously longitudinally furrowed leaves, and also the fruits are not to be confused with any already described. The two species differ from one another markedly in fruit characters: *C. macrosperma* has a very large pyxidium broadest near the mouth, a calycalis zone of which the sepals are nearly suppressed and distinct from one another, a broad suberect interzonal band, and oblong-lanceolate seeds five times as long as broad; *C. Krukovii* has a smaller pyxidium contracted at the mouth, a calycalis zone of which the sepals are comparatively large and interconnected, a narrow flat interzonal band, and obovate-oblong seeds three times as long as broad. The pyxidium of *C. glabra* is the same shape as that of *C. macrosperma;* the seeds of *C. glabra* resemble those of *C. Krukovii.*

CARINIANA MICRANTHA Ducke. Matto Grosso: near Tabajara, upper Machado River region, *Krukoff 1439*. The local name "Tauary" is noted by the collector, whose specimens are from a tree 35 meters tall and 1.5 meters in diameter near the base. The wood is noted as very hard, it having taken two men one day to fell the tree.

CARINIANA KUHLMANNII Ducke. Matto Grosso: near Tabajara, upper

Machado River region, *Krukoff 1375*. A second collection, not far from the type locality. The name "Tauary" is also applied to this tree, according to Mr. Krukoff.

### MELASTOMATACEAE

**Loreya strigosa** Gleason, sp. nov. Arbor 12 m. alta; rami floriferi cinerei glabri, superiores foliosi dense villosi, pilis curvatis brunneis basi incrassatis 3 mm. longis; folia ternata in quoque verticillo satis inaequalia, petiolo villoso 5–8 mm. longo, membranacea oblongo-lanceolata abrupte acuta vel subacuminata, infra medium longe cuneata irregulariter denticulata vel subintegra et quasi ciliata, nervis marginalibus tenuibus praetermissis 3-pli-nervia, supra parce scabro-hirsuta et ad costam strigosa, subtus hirsutula, venis supra planis et obscuris, subtus prominentibus, secundariis sub angulo 70° orientibus, tertiariis atris reticulatis; fasciculae ternatae ad axillas foliorum delapsorum, quaque sub-10-flora; pedicelli 3–7 mm. longi dense strigosi; hypanthium campanulatum, 4.8 mm. longum dense strigosum, pilis subulatis incurvis circiter 1 mm. longis; calycis tubus erectus 2.2 mm. longus sicut hypanthium strigosus; sepala semicircularia e sinibus angustis acutis 1.8 mm. longa, extra breviter strigosa vel ad marginem subglabra, intus sericea; petala 5 ovato-oblonga 10–11 mm. longa 4 mm. lata obtusa, extra ad basin sericea ceterum strigosa; stamina 10 isomorpha; filamenta crassa complanata glabra 7 mm. longa ad apicem sensim dilatata; antherae oblongae 6.5 mm. longae obtusae 4-loculares poris 2 minutis terminalibus dehiscentes, connectivo simplici; ovarium toto inferum 5-loculare, ovulis numerosis, summo concavum; stylus glaber sulcatus 7 mm. longus, stigmate ovoideo.

Type, *Krukoff 1593*, collected Dec. 21, 1931, in a very old clearing near the source of the Jatuarana River, State of Matto Grosso. The vernacular name is "Jambo."

The genera *Bellucia* and *Loreya* are weakly separated. According to Cogniaux the former bears axillary flowers and the stamens are 2-pored, while the latter bears flowers on the old wood and the stamens are 1-pored. There is also a distinct difference in the general aspect of the plant, depending primarily on pubescence, leaf-shape, and character of the hypanthium. In bud the petals of *Loreya* are imbricated in such a way that they appear acute and at once suggest the related genus *Henriettella*. The anthers are distinctly 1-pored in *Loreya mespiloides*, the second species to be described, and possibly in other species as well. In *Loreya ovata* they are distinctly 2-pored; so are they in certain Peruvian and Bolivian specimens of recent collection which have been tentatively referred to *Loreya Spruceana*, and they are apparently 2-pored in Spruce's type of the latter as well.

Within the genus, *L. strigosa* is obviously related to *L. Spruceana*, but differs from it in its hirsute stems, its narrower hirsute ternate leaves, its

strongly strigose hypanthium, its longer sepals, and its narrower strigose petals. The size of the leaves varies from 36–61 cm. long by 8–18 cm. wide; the lateral nerves arise 4–12 cm. from the base of the petiole.

**Loreya quadrifolia** Gleason, sp. nov. Arbor 8 m. alta; rami foliiferi dense villosi, pilis brunneis 3–5 mm. longis patentibus vel sursum curvatis; rami defoliati glabrati; petioli sicut rami villosi, 3–5 mm. longi; laminae tenues, oblongo-oblanceolatae, 15–22 cm. longae, 5–8 cm. latae, subiter acuminatae, margine paullum undulatae, a medio ad basin angustam sensim cuneatae, utrinque pilosae, pilis subrigidis circiter 2 mm. longis, infra ad costam densius pilosae pilis longioribus, 3-pli-nerviae, nervis lateralibus 3–5 cm. supra basin orientibus, venis omnibus supra obscuris, subtus paullum elevatis, secundariis sub angulo circiter 60° adscendentibus, inter se 6–8 mm. dissitis; flores 5-meri subsessiles ad nodos defoliatos dense fasciculati; hypanthium late campanulatum, 5 mm. longum, densissime hirsutum, pilis 2–4 mm. longis, patentibus vel subrecurvatis, e basibus contiguis papillosis; calycis tubus 2.8 mm. longus, suberectus; sepala late triangulari-semicircularia, 2.2 mm. longa, 5 mm. lata; calyx extra ubique hirsutus hypanthio similis, intus densissime sericeus; petala ad anthesin patentia, triangulari-lanceolata, 11–12 mm. longa, 4–4.5 mm. lata, acuta, ad basin tenuia glabra, superne incrassata extra ad medium dense strigosa marginibus glabra inflexa; stamina isomorpha; filamenta 6.7 mm. longa, apicem versus incrassata; antherae crassae oblongae 4-loculares, poris 2 minutis dehiscentibus, connectivo elevato nec appendiculato nec producto; ovarium inferum parvum 5-loculari, ovulis numerosis; stylus 19 mm. longus, superne sensim dilatatus, stigmate ovoideo 2 mm. longo.

Type, *Krukoff 1510*, collected Dec. 3, 1931, on "terra firma" near Tabajara, upper Machado River region, State of Matto Grosso. *L. quadrifolia* is undoubtedly closely related to *L. strigosa*, described above, differing from it in its leaves in whorls of four, its longer, stiffer, and more spreading pubescence, its hirsute hypanthium and calyx, and its much longer style.

## MYRSINACEAE

**Weigeltia glomerulata** A. C. Smith, sp. nov. Arbor 10 m. alta, trunco 7–8 cm. diametro; ramulis teretibus, cortice cinereo striato juventute densissime fusco-pulverulento mox glabrescente; petiolis plerumque glabris striatis leviter canaliculatis distaliter anguste alatis 15–30 mm. longis; laminis papyraceis viridibus glabris densissime minute pellucido-punctatis, ovatis vel ellipticoovatis, 15–25 cm. longis, 6–12 cm. latis, basi attenuatis, apice subacutis vel obtusis, margine integris et cartilagineis, costa supra impressa subtus prominente, nervis lateralibus primariis 8–11 in quoque latere arcuato-adscendentibus supra planis subtus prominentibus, venulis copiose reticulatis; inflorescentiis axillaribus vel e ramulis infra folia breviter stipitatis, subspicatis vel anguste paniculatis, 2–9 cm. longis, ramulis minute ferrugineo-pulverulentis;

bracteis deciduis lanceolatis 2 mm. longis extra pulverulentis; floribus subglabris 4-meris sessilibus in glomerulis aggregatis; sepalis membranaceis plerumque epunctatis ovatis rotundatis circiter 1 mm. longis et latis; petalis velut sepalis sed leviter majoribus, basi connatis; staminibus quam petalis brevioribus, filamentis ad 0.3 mm. longis, antheris subglobosis circiter 0.5 mm. diametro, apice rotundatis, basi cordatis, per rimas longas dehiscentibus; ovario minuto conico glabro, stylo brevi; fructu nigrescente globoso ad 4 mm. diametro, stylo persistente coronato.

Type, *Krukoff 1144*, collected Sept. 16, 1931, in high forest drained by the Cupary River, on the plateau between the Xingu and Tapajos Rivers, State of Pará. It is a species of the Section *Euweigeltia* Mez, in which it is related to *W. Schomburgkiana* Mez. The longer petioles, broader leaves, and narrower inflorescences distinguish *W. glomerulata* from the Guiana species. The flowers of the new species (on our specimen not fully mature) are unusually minute; the filaments are shorter than the anthers, while *W. Schomburgkiana* has them 2–4 times as long.

**Weigeltia multiflora** A. C. Smith, sp. nov. Frutex dioicus gracilis erectus 0.5 m. altus, cortice fusco; foliis comatis, petiolis crassis ad 1 cm. longis fusco-puberulis, laminis chartaceis olivaceis obovatis, 45–65 cm. longis, 10–16 cm. latis, basi longe attenuatis, apice acutis et mucronatis, margine integris saepe undulatis, utrinque (praecipue subtus) lineolatim striatis et parce ferrugineo-pulverulentis, supra saepe glabris, costa utrinque prominente supra canaliculata, nervis lateralibus primariis 15–18 in quoque latere arcuato-adscendentibus utrinque elevatis, venulis copiosissime reticulatis utrinque leviter elevatis, ramulis ultimis liberis; inflorescentiis ♂ axillaribus paniculatis stipitatis multifloris, ad 25 cm. longis et 8 cm. latis, ferrugineo-pulverulentis, ramulis bracteis papyraceis puberulis oblongis acutis 6–10 mm. longis subtentis; floribus 3-meris basi decidue bracteolatis; pedicellis velut ramulis pulverulentis, 2.5–4 mm. longis; sepalis vix ultra ⅓ connatis anguste imbricatis ovatis rotundatis, 1–1.2 mm. longis et latis, punctis magnis nigrescentibus pictis, margine membranaceis et arcte fimbriatis; petalis velut sepalis pictis oblongis, 4.5–5 mm. longis, 2–2.5 mm. latis, apice obtusis, margine glabris, maturitate recurvatis; staminibus quam petalis brevioribus prope basin insertis, filamentis carnosis 2 mm. longis apice gracilibus, antheris subglobosis 0.6–0.8 mm. diametro, dorso glandulosis, apice rotundatis, basi cordatis, per rimas dehiscentibus; ovarii rudimento minuto globoso ferrugineo-pulverulento; floribus ♀ ignotis.

Type, *Krukoff 1388*, collected Nov. 23, 1931, on "terra firma" near Tabajara, upper Machado River region, State of Matto Grosso. Another collection from the same region is *Krukoff 1377*. It is a species of the Section *Triadophora* Mez, based on the Colombian *W. Schlimii* (Hook. f.)

Mez. In common these plants have 3-merous flowers and extraordinarily robust leaves and inflorescences, together with noticeable subepidermal sclerenchyma fibers. Their relationship with other species of *Weigeltia* does not seem very close. Compared with *W. Schlimii*, *W. multiflora* has a simpler habit, the leaves entire rather than toothed, and the inflorescence simply rather than twice pinnate. In flower structure there is little difference, although the new species has narrower petals and strictly glabrous anthers.

SAPOTACEAE

**Lucuma inflexa** A. C. Smith, sp. nov. Arbor mediocris; ramulis teretibus fuscis vel cinereis, juventute ferrugineo-sericeis mox glabrescentibus; petiolis mox glabris rugosis canaliculatis, 7–12 mm. longis, basi nigrescentibus et incrassatis; laminis chartaceis glabris oblongis, 8–12 cm. longis, 3–5 cm. latis, basi cuneatis, apice acuminatis (apice ipso obtuso), margine integris, nervis lateralibus primariis 7–9 in quoque latere arcuato-adscendentibus, cum costa supra subplanis subtus elevatis, venulis reticulatis subtus prominulis; inflorescentiis in nodis fasciculatis 2–5-floris; pedicellis gracilibus 25–40 mm. longis, superne incrassatis, densissime ferrugineo-sericeis; calyce velut pedicellis extra sericeo, intus minutissime pulverulento, sepalis 5 late imbricatis inaequalibus, exterioribus carnosis deltoideo-ovatis, circiter 4 mm. longis et 5 mm. latis, interioribus submembranaceis oblongis, circiter 3 mm. latis; corolla campanulata tenuiter carnosa extra sericea intus glabra 5–6 mm. longa, lobis 5 tubum subaequantibus ovatis, 3–3.5 mm. latis, apice rotundatis; staminodiis late ovatis inflexis, 1.5 mm. longis et latis, apice obtusis; staminibus (saepe anantheris) basi loborum insertis, filamentis carnosis minutis (0.5 mm. longis), antheris valde inflexis ovoideis, 2 mm. longis, 1 mm. latis, 4-alatis, apice apiculatis, per rimas extrorsas dehiscentibus; ovario dense sericeo depresso-globoso plus minusve 6-lobato, loculis 3, ovulo 1 in quoque loculo; stylo tereti ovarium aequante, apice minute 3-lobato.

Type, *Krukoff 1505*, collected Dec. 3, 1931, on "terra firma" near Tabajara, upper Machado River region, State of Matto Grosso. The plant is locally known as "Cramary;" the inner bark is said to yield a white latex. It is a species of the Section *Rivicoa* A. DC., related to *L. retusa* Spruce and *L. obscura* Huber. With the latter it has in common a 3-celled ovary. *L. inflexa*, however, has the leaves somewhat smaller and the pedicels longer than *L. obscura*. The staminodes of the present species are obtuse and the anthers practically sessile, in which points it also differs from *L. obscura*. Flowers in which the anthers are completely lacking are very common, many of the closed buds which were dissected showing this feature. The specific name refers to the strongly inflexed anthers.

**Lucuma platyphylla** A. C. Smith, sp. nov. Arbor 20 m. alta, trunco 25–30 cm. diametro; ramulis subteretibus crassis densissime ferrugineo-velutino-

tomentosis demum glabrescentibus; petiolis tomentosis crassis leviter canaliculatis, 15–40 mm. longis; laminis chartaceis bullatis obovato-oblongis, 18–30 cm. longis, 8–12 cm. latis, basi acutis, apice caudato-acuminatis vel cuspidatis, margine integris et leviter revolutis, supra praeter costam glabris, subtus pilis ferrugineis erectis stipitatis 2–4-ramosis 0.5 mm. longis pubescentibus, costa supra subplana subtus prominentissima, nervis lateralibus 13–16 in quoque latere patulis, prope margines anastomosantibus, supra impressis subtus prominentibus, venulis copiosissime reticulatis supra prominulis subtus elevatis; floribus 3–8 in fasciculis ad nodos infra folia; pedicellis gracilibus 12–16 mm. longis, densissime ferrugineo-tomentosis; sepalis 5 uniseriatis basi connatis deltoideo-ovatis, circiter 3 mm. longis et latis, apice acutis, extra velut pedicellis tomentosis, intus minute flavescenti-sericeis; corolla submembranacea flavida glabra circiter 6 mm. longa, lobis 5 tubum aequantibus ovatis rotundatis 2.5–3 mm. latis, margine saepe sinuatis; staminodiis carnosis ovatis, circiter 2.5 mm. longis et 1.5 mm. latis, apice acuminatis; filamentis fertilibus carnosis 1–1.5 mm. longis, antheris inflexis ovoideis, 2 mm. longis, 1.2 mm. latis, basi cordatis, apice obtusis; ovario depresso-globoso circiter 2 mm. diametro, dense tomentoso (pilis ad 0.7 mm. longis), valde 5-costato, loculo unico non centrali, ovulo unico; stylo quam ovario breviore, minute lobato.

Type, *Krukoff 1316*, collected Nov. 12, 1931, on "terra firma" near Tabajara, upper Machado River region, State of Matto Grosso. The plant is locally known as "Abiurana;" the inner bark yields a white latex. It is a species related to *L. ramiflora* (Mart.) A. DC., which it resembles in habit, pubescence, and inflorescence. However, *L. platyphylla* has the leaves much larger, the flowers 5- rather than 4-merous, and the ovary 1- rather than 2-celled. The numerous ovaries I have dissected show only one locule containing one ovule; the single locule is not central, and the distinctly 5-costate ovary indicates that the other 4-locules are abortive.

**Lucuma anibaefolia** A. C. Smith, sp. nov. Arbor glabra 10 m. alta, trunco 15 cm. diametro; ramulis teretibus striatis, juventute stramineis; petiolis saepe violaceis canaliculatis, 6–12 mm. longis; laminis olivaceis chartaceis oblongis, 10–17 cm. longis, 2.5–5 cm. latis, basi subattenuatis, apice obtuse acuminatis, margine integris saepe sinuatis, costa utrinque prominente, nervis lateralibus primariis 12–14 in quoque latere arcuato-adscendentibus supra subplanis subtus elevatis, venulis copiosissime reticulatis utrinque prominulis; inflorescentiis 5–10-floris, floribus fasciculatis, pedicellatis (pedicellis gracilibus 3–5 mm. longis, flavescenti-puberulis), basi bracteatis, bracteis deltoideis acutis ad 1 mm. longis; calyce extra puberulo intus glabro, sepalis 6 imbricatis subaequalibus ovatis obtusis, 1–1.5 mm. longis et latis, margine membranaceis et fimbriatis; corolla submembranacea glabra breviter campanulata 2 mm. longa, lobis 6 tubum subaequantibus ovatis obtusis 1 mm. latis, margine fimbriatis; staminodiis ovatis inflexis, circiter 0.6 mm. longis, apice acutis;

staminibus prope basin tubi adnatis, filamentis 0.3 mm. longis, antheris ovoideis, circiter 0.5 mm. longis, parcissime puberulis, per rimas laterales dehiscentibus; ovario depresso-globoso, sub anthesi 1 mm. diametro, minutissime sericeo, loculo unico, ovulo unico; stylo carnoso, circiter 0.7 mm. longo, apice truncato.

Type, *Krukoff 1447*, collected Nov. 25, 1931, on varzea land along river, near Tabajara, upper Machado River region, State of Matto Grosso. A local name is "Abiuhy." It is a species probably best placed in the Section *Eremoluma* (Baill.) Engl., because of its 1-celled 1-ovuled ovary, although 6-merous flowers have not previously been noted in the section. It is related to *L. rostrata* Huber, but has proportionately narrower leaves, a more compact inflorescence, and much smaller flowers than that species.

### EBENACEAE

**Diospyros Melinoni** (Hiern) A. C. Smith, comb. nov. *Maba Melinoni* Hiern, Trans. Cambr. Phil. Soc. 12: 143. 1873.

Examination of an isotype of this species in the herbarium of the New York Botanical Garden indicates that the ovary is habitually 8-celled and the styles 4. It belongs, therefore, in the genus *Diospyros*, Section *Rospidios* Hiern. Its nearest relative is apparently *D. glomerata* Spruce. In leaf texture and flower structure the two species are very similar, but *D. Melinoni* has leaves only half as large, the flowers less densely aggregated, and the floral pubescence sparser. *D. glomerata* has the corolla densely sericeous rather than essentially glabrous.

The species has been collected by Krukoff (no. *1188*) in high forest drained by a tributary of the Cupary River, on the plateau between the Xingu and Tapajos Rivers, State of Pará. As this specimen bears ♂ flowers, the following description may be appended to the original:

♂ inflorescence axillary, short, 2–6-flowered, the peduncles up to 3 mm. long; flowers subsessile, subtended by sericeous oblong bracts about 2 mm. long; calyx flavo-sericeous on both sides, about 5 mm. long including lobes, the lobes 5 or 6, deltoid, acute, 2.5–3 mm. long, about 1.5 mm. broad; corolla white, rotate, 5- or 6-lobed nearly to the base, the lobes thin-carnose, obovate, 5–6 mm. long, 3–4 mm. broad, rounded at apex and frequently recurved, sericeous towards the base without, glabrous within; stamens 45–55, lightly coherent at base, the filaments 0.5–0.8 mm. long, setose with hairs about 1 mm. long, the anthers oblong-linear, acute, curved, about 2 mm. long, setose at apices and on connectives; ovary apparently completely lacking.

**Diospyros Krukovii** A. C. Smith, sp. nov. Frutex 3–4 m. altus; ramulis teretibus cinereo-pulverulentis demum glabrescentibus, internodiis 2.5–4.5

cm. longis; petiolis canaliculatis 7–12 mm. longis, velut ramulis decidue pulverulentis; laminis chartaceis siccitate fuscis oblongis, 15–25 cm. longis, 5.5–8 cm. latis, basi rotundatis et abrupte cuneatis, apice longe acuminatis (apice ipso obtuso), margine integris et leviter recurvatis, utrinque glabris (vel subtus juventute parce flavescenti-strigosis), nervis lateralibus 8–10 in quoque latere arcuato-adscendentibus, cum costa supra elevatis subtus prominentibus, venulis copiose reticulatis utrinque leviter elevatis; floribus ut videtur semper hermaphroditis axillaribus solitariis subsessilibus, basi bracteatis, bracteis 3–5 ovatis, 5–6 mm. longis, 4 mm. latis, extra strigosis intus glabris; calyce ubique dense flavescenti-sericeo, fere ad basin 5-lobato, lobis deltoideo-ovatis acutis, 6–7 mm. longis, 3–4 mm. latis; corolla subrotata alba tenuiter carnosa mox decidua profunde 5-lobata, lobis prope bases imbricatis ovato-oblongis subacutis, 8–10 mm. longis, 4–5 mm. latis, extra lineis latis pilorum pallidorum vestitis, intus glabris; staminibus uniseriatis circiter 16 distinctis, ad basin corollae adfixis; filamentis stramineis glabris carnosis circiter 1 mm. longis; antheris oblongo-linearibus acutis 5 mm. longis, connectivis pilos pallidos setosos ad 1.5 mm. longos gerentibus; ovario globoso densissime strigoso (pilis 2 mm. longis); stylis 3 basi connatis, 3.5 mm. longis, divaricatis, stigmatibus in lobis 3 planis irregularibus breviter fimbriatis divisis; loculis 6, ovulo 1 in quoque loculo; fructibus globosis, maturitate 2.5–3.5 cm. diametro, persistenter strigosis, pericarpio coriaceo, seminibus maturitate 2 vel 3 ovalibus circiter 15 mm. longis et 8 mm. latis.

Type, *Krukoff 1564*, collected Dec. 7, 1931, on "terra firma" near Angustura, near the source of the Jatuarana River, Machado River region, State of Matto Grosso. According to Hiern's monograph, *D. Krukovii* would fall into the genus *Maba*, Section *Tricanthera*, by virtue of its 5-merous flowers and 6-celled ovary. In this section, it resembles *M. sericea* (A. DC.) Hiern in its solitary hermaphrodite flowers. Two recent students[1] have considered *M. sericea* best placed in *Diospyros*, where it constitutes a rather anomalous species in the Section *Rospidios* Hiern. This treatment is here followed.

From the species of this alliance, *D. sericea* A. DC., *D. pseudoxylopia* Mildbr., and *D. dichroa* Sandwith, the new species is readily distinguished by having its leaves fully twice as large and essentially glabrous beneath, as well as by floristic details. *D. Krukovii* apparently always has the flowers perfect, whereas the above mentioned species are said to be dioecious or polygamo-dioecious. The fact that the subequal stamens are attached to the corolla in a single row is also a point upon which *D. Krukovii* agrees with neither *Maba* nor *Diospyros*, as they are commonly understood.

[1] Mildbr., Notizbl. **10**: 195. 1927; Sandwith, Kew Bull. **1931**: 483. 1931.

## APOCYNACEAE

**Prestonia Lindleyana** Woodson, nom. nov. *Haemadictyon calycinum* Lindl.; Miers, Apoc. So. Am. 259. 1878; not *Prestonia calycina* Muell. Arg. (1860).

Matto Grosso: Near Tabajara, upper Machado River region, *Krukoff 1427.*

**Prestonia trifida** (Poeppig) Woodson, comb. nov. *Haemadictyon trifidum* Poeppig; P. & E. Nov. Gen. & Sp. **3**: 67. *pl. 275.* 1845.

Matto Grosso: Near source of the Jatuarana River, Machado River region, *Krukoff 1545.*

**Odontadenia Hoffmannseggiana** (Steud.) Woodson, comb. nov. *Echites grandiflora* G. F. W. Mey. Fl. Esseq. 131. 1818; not Roxb., Roth, Stadelm., or Hoffmsg. *Echites macrantha* R. & S. Syst. 4: 795. 1819; not Spreng. *Echites Hoffmannseggiana* Steud. Nom. ed. 2. **1**: 539. 1840. *Odontadenia speciosa* Benth.; Hook. Journ. Bot. **3**: 242. 1841.

Matto Grosso: Near Tabajara, upper Machado River region, *Krukoff 1457, 1471.*

## VERBENACEAE

**Aegiphila villosissima** Moldenke, sp. nov. Frutex scandens; caulibus et ramis crassis medullosis obtuse tetragonis densissime longeque villosis, pilis fuscis vel ferrugineis patentibus vel reflexis 4–8 mm. longis; ramulis gracilioribus; internodiis valde elongatis 9–18 cm. longis; foliis oppositis; petiolis crassis 5–10 mm. longis dense longeque villosis; laminis laete viridibus membranaceis ovatis vel ovato-ellipticis 11.5–25 cm. longis, 4.5–12 cm. latis, ad apicem abrupte breviterque acuminatis, margine integris et longe ciliatis (pilis subsetosis) saepe subrevolutis, ad basin insigniter rotundatis, utrinque dense villosis, pilis fuscis subsetosis circa 3 mm. longis et plerumque usque ad 1 mm. dissitis; costa supra plusminus impressa, subtus perprominente, utrinque dense villosa praecipue subtus; venis secundariis gracilibus 10–12 in quoque latero arcuato-adscendentibus insigniter 5–17 mm. a margine anastomosantibus dense villosis; inflorescentiis axillaribus terminalibusque paniculatis; panicula terminali bracteata circa 10 cm. longa et 5.5 mc. lata 5-cymifera, ubique dense longeque villosa; paniculis axillaribus longe pedunculatis paniculae terminali consimilibus vel parvioribus et subcymosis; cymis conspicue bracteolatis; pedunculis crassiusculis 2.5–6 cm. longis, ubique sicut ramulis dense longeque villosis; bracteis paucis lanceolatis circa 1 cm. longis et 4 mm. latis, ad apicem et basin attenuatis, utrinque densissime longeque villosis; bracteolis linearibus elongatis usque ad 17 mm. longis dense villosis; calyce levi herbaceo circa 1.5 mm. longo et 3.3 mm. lato patentissimo e basi campanulato, extra dense longeque villoso, intus glabro, margine 4-lobato, lobis brevi-

bus obtusis; corolla infundibulariformi, tubo (immaturo?) anguste cylindrico circa 1.5 mm. longo glabro, lobis 4 oblongo-lingulatis 3.8–4 mm. longis circa 2.5 mm. latis obtusis glabris; staminibus 4 ad oram tubi corollae insertis; filamentis (immaturis?) filiformibus circa 0.7 mm. longis, inferne pilosis; antheris oblongis circa 1.3 mm. longis et 0.7 mm. latis; stylo capillari circa 2.6 mm. longo glabro; ramulis stigmatis bifidi circa 2 mm. longis valde patentibus; ovario depresso-oblongo circa 1 mm. longo et 1.3 mm. lato, ad apicem umbilicato et 4-sulcato, 4-loculare.

Type, *Krukoff 1400*, collected Nov. 23, 1931, on "terra firma" near Tabajara, upper Machado River region, State of Matto Grosso. The species is most closely related to the Peruvian *A. cordata* Poepp. The latter, however, differs in its much smaller and distinctly subsessile leaves whose blades are conspicuously cordate or subcordate at the base and in its short-pedunculate or subsessile cymes and lack of axillary panicles. Its flowers, also, are much larger than in our species, but in ours the flowers are not yet quite open, so that their dimensions may be unreliable.

## SOLANACEAE

**Solanum quaesitum** Morton, sp. nov. Subg. *Leptostemonum*, Sect. *Oliganthes;* frutex erectus ca. 4 m. altus; rami superiores robusti, ca. 6 mm. diametro, leviter sulcati, pilis stellatis subferrugineis longe stipitatis crebris obtecti, aculeis recurvatis ca. 5 mm. longis, ca. 3 mm. basi latis, apice acutis, parce pubescentibus armati; folia superiora solitaria; petioli pro magnitudine laminae sat longi, ca. 32 mm. longi, dense pubescentes, pilis stellatis longe stipitatis vestiti, aculeati, aculeis rectis usque 1 cm. longis basi perspicue pubescentibus; laminae ovatae, usque ad 23 cm. longae et 17 cm. latae, vix vel evidenter 5 lobatae, basi inaequaliter rotundatae, in petiolum haud decurrentes, apice acutiusculae, membranaceae, utrinque (in statu sicco) olivaceae, supra pilis stellatis pauciradiatis longe stipitatis (cum pilis minoribus simplicibus apice glanduliferis intermixtis) crebre vestitae, venis lateralibus primariis ca. 5, supra paullum impressis, subtus prominentibus; inflorescentiae extraaxillares, a foliis remotae, ca. 8-florae; pedunculus communis ca. 5 cm. longus; pedicelli ca. 15 mm. longi, densissime pubescentes, pilis longissime stipitatis; calyx 5-partitus, lobis reflexis, lanceolatis, ca. 17 mm. longis, coriaceis, margine membranaceis erosis, intus glabris, extus in tota superficie densissime pubescentibus, pilis stellatis, pauciradiatis, longissime stipitatis, stipitibus robustis usque 5 mm. longis, lutescentibus; corolla violacea (sec. Krukoff), rotata, magna, explanata ca. 5 cm. lata, plicata, 5-lobata, lobis lanceolatis acutis, extus dense tomentosis (pilis stellatis vix stipitatis), intus in vena media parce pubescentibus, fere usque ad apices membranis interpetalariis glabris conjunctis; filamenta ca. 2 mm. longa, glabra; antherae lineari-lanceolatae, ca. 13 mm. longae, 1.5 mm. latae, poris apicalibus minutis; ovarium subglobosum, glabrum; stylus rectus, stamina fere aequans, glaber; pedicelli fruc-

tiferi ca. 3 mm. diametro; calyx fructiferus auctus, ca. 2.5 cm. latus, valde et irregulariter incrassatus, setis sparsis persistentibus; bacca globosa, ca. 4 cm. diametro, glabrata; semina obovoidea vel suborbicularia, ca. 3 mm. diametro, testa favosa.

Type, *Krukoff 1637*, collected Dec. 20, 1931, in a new clearing near the source of the Jatuarana River, Machado River region, State of Matto Grosso. Closely allied to *Solanum setosicalyx* Rusby, which differs in the indument of the upper leaf surface being scabrous and composed of numerous stout simple hairs, rather than stellate-tomentose and glandular as in the present species. Rusby in his description of *S. setosicalyx* does not indicate the relationship of the species even as to the section. Examination of a specimen of the type collection[1] in the U. S. National Herbarium shows, however, that it belongs to the section *Oliganthes*, and is near or probably synonymous with the earlier *Solanum Vanheurckii* Muell. Arg.,[2] known to me from the original description only.

**Solanum placitum** Morton, sp. nov. Subg. *Pachystemonum*, Sect. *Anthoresis;* frutex erectus, ca. 6 m. altus; rami juniores angulati, scabriusculi, pilis minutis ochraceis stellatis sessilibus vel breviter stipitatis pleniradiatis sat densis praediti; folia solitaria; folia minora axillaria ("auriculae") desunt; petioli foliorum inferiorum usque 3 cm. longi, superiorum subnulli, pubescentia eae ramulorum simili; laminae obovatae, magnae, maximae saltem 34 cm. longae et 16.5 cm. latae, integrae, apice breviter et acute acuminatae, basi longe cuneatae et in petiolum decurrentes, membranaceae, utrinque (in statu sicco) olivaceae, concolores, supra juventute parce strigosae, pilis pro parte maxima simplicibus subulatis basi inflatis apice acicularibus usque 0.25 mm. longis, ca. 0.5 mm. distantibus, pro parte minima (praecipue in venis mediis et lateralibus) stellatis minutis sessilibus pauciradiatis, demum glabratae, tunc pilis 1–3 mm. distantibus, subtus persistente stellato-puberulentae, pilis minutis, sessilibus pauciradiatis, 0.25–0.5 mm. distantibus, venis lateralibus primariis ca. 7, subtus elevatis; inflorescentiae terminales pauciflorae; pedunculus ca. 6 cm. longus, apice furcatus, sicut ramuli pubescens; pedicelli ca. 5 mm. longi, dense stellato-tomentosi, apice non incrassati; calyx subturbinatus, 5-dentatus, ca. 4 mm. longus, 7 mm. latus, extus densissime stellato-tomentosus, pilis minutis sessilibus vel brevissime stipitatis, intus glaber (pilis paucis apicem versus exceptis), venulosus, dentibus deltoideis aequalibus ca. 2 mm. longis, ca. 2 mm. basi latis, apice acutis; corolla alba (sec. Krukoff), rotata, explanata, ca. 17 mm. lata, 5-fida, lobis ovalibus ca. 6 mm. longis apice acutis, extus in tota superficie dense stellato-tomentosis, intus glabris, tubo brevi, ca. 2 mm. longo, glabro; filamenta ca. 2 mm. longa, glabra, basi in

[1] Huachi, head of Beni River, Bolivia, *H. H. Rusby* (Mulford Amazon Expedition) *451.*

[2] Van Heurck, Obs. Bot. 85. 1870.

tubum saltem 0.5 mm. altum connata; antherae late ellipticae, ca. 3 mm. longae, 2 mm. latae, poris magnis introrsis; ovarium globosum, dense stellato-hirsutum; stylus curvatus, ca. 7 mm. longus, 0.6 mm. crassus, pilis paucis stellatis instructus; fructus deest.

Type, *Krukoff 1583*, collected Dec. 20, 1931, in a very old clearing near the source of the Jatuarana River, Machado River region, State of Matto Grosso. The present plant does not appear to be closely related to any of the previously described species of the section *Anthoresis*. It belongs to the general group of species related to *S. verbascifolium* L. The character of the pubescence of the upper surface of the leaves, however, is characteristic, as are also the monadelphous stamens and the shape of the leaves.

## Rubiaceae

**Coussarea machadoana** Standl., sp. nov. Arbor 14-metralis, trunco 30 cm. diam., praeter inflorescentiam omnino glabra, ramulis crassiusculis rectis subteretibus, internodiis elongatis; stipulae 4–5 mm. longae in vaginam brevem incrassatam persistentem connatae, lobis brevissimis apiculatis; folia maxima breviter petiolata coriacea in sicco flavescentia, petiolo crasso 1–1.6 cm. longo; lamina anguste elliptico-oblonga c. 28 cm. longa et 9 cm. lata apice obtusa at 1 cm. longe caudato-cuspidata, acumine obtuso, basi acuta, costa supra elevata, nervis prominentibus, subtus paullo pallidior, costa valde elevata, nervis lateralibus utroque latere c. 14 prominentibus angulo fere recto divergentibus marginem attingentibus leviter arcuatis, venulis vix prominulis reticulatis, margine plus minusve revoluto; inflorescentia terminalis cymoso-paniculata sessilis basi trichotoma dense multiflora 9 cm. longa 14 cm. lata, ramis rectis crassiusculis minutissime pruinoso-puberulis, floribus aggregatis cymulosis vel interdum subumbellatis crasse 1–2 mm. longe pedicellatis, bracteis obsoletis; hypanthium globoso-obovoideum 1–1.5 mm. longum pruinoso-puberulum, calyce truncato 1 mm. alto 2 mm. lato; corolla alba extus glabra in alabastro apice acuta, tubo crassiusculo 8–9 mm. longo apicem versus angustato, lobis 4 lineari-lanceolatis 5–6 mm. longis apicem angustum versus sensim attenuatis intus minute puberulis patentibus; antherae inclusae; stylus gracilis rectus glaber 2.5–3 mm. longus, ramis c. 1 mm. longis.

Type, *Krukoff 1361*, collected Nov. 15, 1931, on "terra firma" near Tabajara, upper Machado River region, State of Matto Grosso. The species is a well-marked one because of its remarkably large leaves, much greater in dimensions than in most members of the genus.

**Elaeagia brasiliensis** Standl., sp. nov. Arbor, ramulis teretibus brunneo-ochraceis crassis, novellis dense puberulis; stipulae ovato-triangulares 6–8 mm. longae persistentes puberulae cuspidato-acuminatae; folia mediocria

coriacea breviter petiolata, petiolo crasso 6–12 mm. longo pilosulo vel glabrato; lamina elliptica vel late elliptica vel rotundato-obovata 8–12 cm. longa 4–9.5 cm. lata obtusa vel apice saepe rotundata basi acuta vel late cuneato-obtusa, supra glabra lucida, subtus fere concolor ad nervos sparse pilosula vel fere omnino glabra in axillis parce barbata, costa elevata, nervis lateralibus utroque latere c. 5 elevatis angulo acuto adscendentibus valde obliquis fere rectis, venulis prominulis arcte reticulatis; inflorescentia terminalis cymoso-paniculata sessilis et e basi trichotoma vel interdum 8 cm. longe pedunculata, dense multiflora, 5–12 cm. longa et aequilata vel latior, ramis brunnescentibus rigidis adscendentibus vel subpatentibus dense minute puberulis, floribus aggregatis sessilibus vel breviter crasse pedicellatis, bracteis deciduis; capsula 4 mm. longa et aequilata obovoideo-globosa dense minute puberula septicidalis, valvis apice saepe breviter bifidis; semina pauca minuta anguste alata, alis laciniatis.

Type, *Krukoff 1018*, collected Sept. 6, 1931, in high forest on "terra firma" at Fordlandia, Tapajos River region, State of Pará. Because of the absence of flowers, there is some uncertainty regarding the generic position of this tree, but the fruit characters and those of the foliage agree better with *Elaeagia* than with any other group. No species of that genus have been reported heretofore from Brazil, although they are to be expected there.

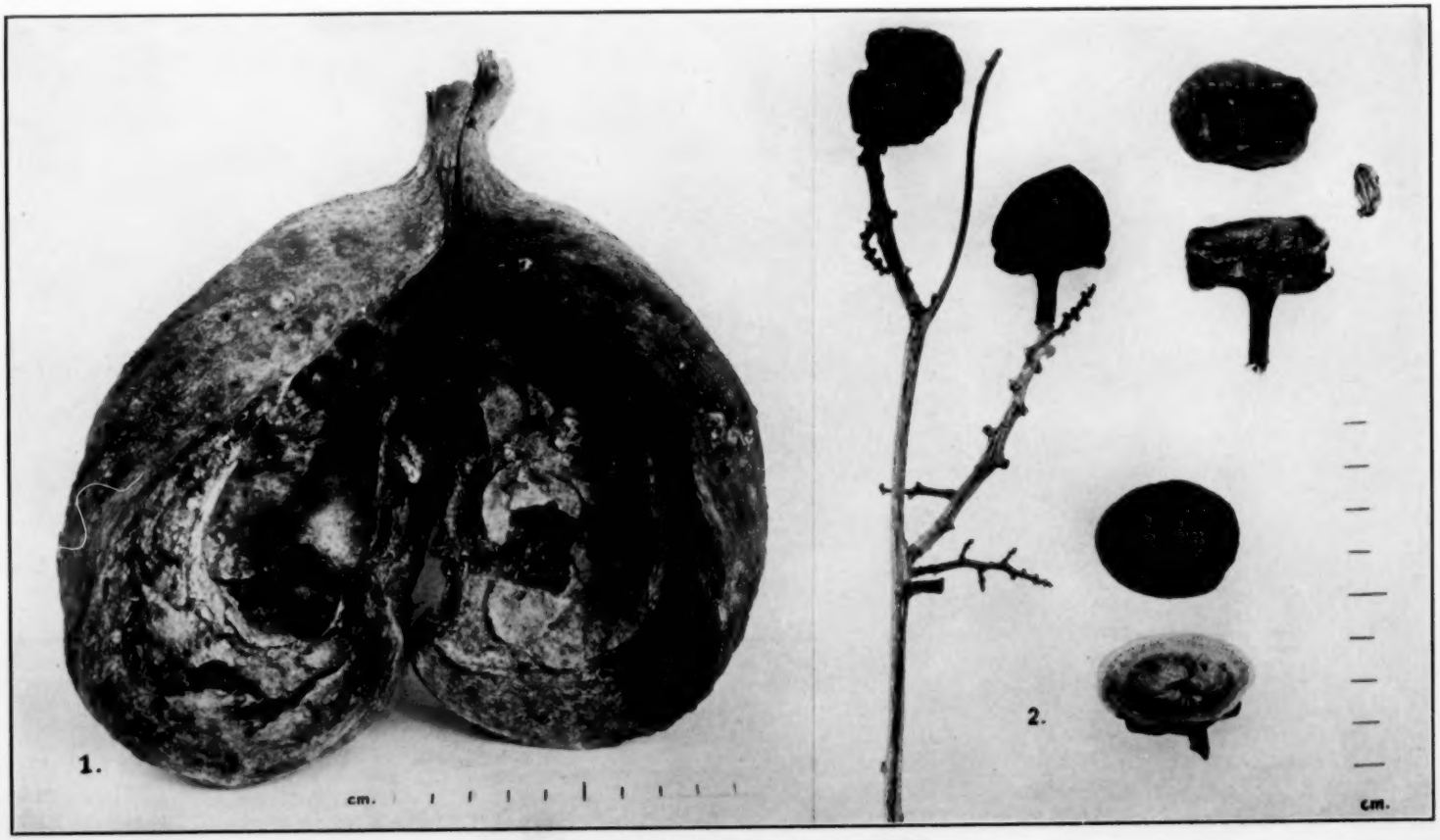
**Explanation of plates 21, 22**

Plate 21

1. Fruit of *Sterculia megalocarpa*.
2. Old inflorescence and fruits of *Eschweilera truncata*.

Plate 22

1. Seed of *Couratari macrosperma*.
2. Pyxidium of *Couratari macrosperma*.
3. Pyxidium of *Couratari Krukovii*.
4. Seed of *Couratari Krukovii*.




GLEASON AND SMITH: STERCULIA: ESCHWEILERA

UNIV. OF MICH.

GLEASON AND SMITH: COURATARI

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# Pollen-tube behavior in Hemerocallis with special reference to incompatibilities

A. B. Stout and Clyde Chandler

(with plate 23 and seven text figures)

In the various daylilies (*Hemerocallis*) seed-production to self-pollination ranges from complete failure due to incompatibility in fertilization to a high degree of self-fertility with many individuals and clons that are only feebly self-compatible and for which seed-production to self- or close-pollination is always low. Quite the same grades of seed-production are also seen in cross-fertilizations. The daylilies are perennials, spreading by crown-branching or by rhizomes to give well-established plants or groups of plants and hence numerous flowers are easily obtained for any plant or for plants of the same clon. The pistils of the flowers are, for certain types, as much as 10.5 cm. in length and fertilizations that are compatible are usually completed in 24 hours or less.

Thus the daylilies afford excellent material for determining pollen-tube growth (1) in various grades of self- and cross-incompatibility, (2) after premature and delayed pollinations and (3) after inter-specific cross-pollinations, and the degree of fruit and seed production following each kind of pollination is readily determined in controls. A very brief mention of the pollen-tube behavior in *Hemerocallis* has been made (Stout, 1931a). The present paper will present the data in detail with graphs for various curves of growth.

## MATERIALS AND METHODS

In obtaining material for study, as a rule as many as 50 flowers of a single clon that had opened under glassine bags were pollinated at one time, usually at or near 9:00 A.M. It is at this time that the anthers of most flowers of the diurnal daylilies are fully dehiscing and the pistils are receptive to compatible fertilizations. At intervals thereafter, at least three flowers were removed and their pistils preserved for study. For premature pollination, delayed pollination and pollination of the flowers of night-blooming sorts, applications of pollen were made at other hours.

In the study of the pollen tubes in the pistils the paraffin method was utilized, but mostly a method of direct dissection and staining was employed as already described in considerable detail (Chandler, 1931; Stout, 1931b). The pistils are preserved in a solution of 100 c.c. of 70% alcohol and 6 to 8 c.c. of commercial formalin (37%). In staining, the pistils are dissected and a few drops of aceto-carmine (saturated solution in 45% acetic acid) are made to flow the entire distance of the style. After a few

seconds a drop of magenta (1% aqueous solution) or a drop of aceto-carmine to which has been added a trace of ferric acetate is placed on the style. The excess stain is removed with absolute alcohol. Glycerine and a cover glass complete a preparation which may be kept for study for a period of several months.

With the use of a microscope the extent of the growth of the pollen tubes in the various pistils is determined and tabulated. If tubes extend into the lower part of the style the ovary is also sectioned and stained. By this method it is rather easy to determine the position of the ends of the pollen tubes, their distribution in the pistil, and their appearance. The pistils of the various kinds of daylilies differ somewhat in length. For example, the styles of the *H. Thunbergii* range from 7 to 8 cm. in length, and those of the *H. fulva* clon Europa range from 9.2 to 9.6 cm. in length. In plotting the extent of pollen-tube growth in these plants the style of a pistil is divided into ten equal units and thus the data for numerous pistils differing somewhat in length may be presented in the same type of graph.

## STRUCTURE OF PISTILS AND COURSE OF POLLEN TUBES

The long slender styles of the pistils of daylilies taper toward the stigma which is slightly expanded into three somewhat definite lobes with the surface covered with elongated papillae (plate 23, figs. 2, 3). Within the style a canal (the "Leitkanal" of Behrens, 1875) extends continuously from the stigmatic surface to the ovary chambers (plate 23, fig. 8). The center of the face of the stigma is somewhat funnel-shaped and the opening into the canal is narrow and almost closed by the interlocking of papillae. A short distance below the stigma the canal increases in diameter and occupies about one-third the diameter of the style (plate 23, figs. 4, 5). For a considerable distance the canal is somewhat three-lobed in the outline of a cross section and there are three narrow strips of irregular cells extending vertically along the wall in the relative position of the placentae, but between these the canal is lined with a layer of somewhat enlarged cells which, however, present a smooth surface. There is no specialized conducting tissue at any point through which pollen tubes travel endotropically. The continuous canal which extends through the style and into the ovary chambers allows any secretions that may be formed to mingle somewhat freely, and it is possible that secretions from the ovary may travel upward to some distance on the wall of the canal.

The stigmatic surface of the pistil of *Hemerocallis* is covered with long slender papillae. In the bud shortly before the flower opens these papillae are erect and turgid. Microscopic examination shows that a

spirally creased layer of cutin covers a thick layer of mucilage which surrounds the cytoplasm of each papillate cell. When the flowers open the papillae spread apart somewhat, especially at their tips. (plate 23, fig. 2). Some of the tips of the papillae become enlarged, the folds become irregular in size bulging in places, and a few small droplets can be seen along the creases of the folds. Soon a thin film of secretion covers the exterior of the papillae thus spreading over the exposed surface. This condition prevails for the time during which pollen is first shed and pollination is accomplished. But soon the papillae near the center of the stigma collapse and the mucilage appears to be liberated near the base of the end cells of the papillae. Toward the end of the day a few papillae around the outer edge of the stigma may still be turgid but the papillae over most of the stigma are collapsed and considerable secretion is released.

Pollen tubes pass over and between the papillae of the stigmatic surface and enter the stylar canal and then travel on into the ovary without being compelled to penetrate through tissues. The ends of the pollen tubes frequently linger in the vicinity of the ovary and in some cases there is later a resumption of growth in this region. Such responses suggest a very direct action of chemotactic substances originating from the ovary or its ovules. In only a few cases here reported was there any swelling of the ends of some of the tubes followed by bursting.

In daylilies large numbers of pollen tubes are usually not present within the styles even when there is a high degree of self-fruitfulness as this is judged by the production of large capsules well filled with seeds. There appears to be a fundamental condition which restricts the number of tubes which enter the stylar canal.

The graphs which follow show the extent to which at least several tubes and usually a number of tubes penetrated at the various time-intervals. In most cases the data for the curves of growth are remarkably consistent.

## DATA ON GROWTH OF POLLEN TUBES

*Pollen-tube growth in Hemerocallis fulva clon Europa.* The fulvous daylily, *H. fulva* clon Europa, is completely self-incompatible (Stout, 1921; 1926). This clon is a triploid with much mixiploidy fluctuating about a chromosome number of 33, (3×11), (Stout, 1932). There is much abortion of pollen but some pollen is viable and functional in fertilizations and for this there is complete self-incompatibility in that capsules do not even start to develop to self-pollination. This clon is strictly diurnal in habit of blooming and all "normal" pollinations were made at or near the hour of 9:00 A.M.

Several thousand self-pollinations and intra-clonal-pollinations have been made for various plants of this clon obtained from many localities in Europe and America. In every case there was *no enlargement* of the ovaries and the flowers promptly abscissed. When inter-specific cross-pollinations are made, using pollen of *H. Thunbergii* or *H. citrina*, the ovaries begin to enlarge and they usually remain attached to the pedicel for at least 10 days, toward the end of which many of the capsules quickly wither and soon fall. Only a very few of such capsules remain to maturity and give viable seeds. Of a total of about 10,000 such cross-pollinations

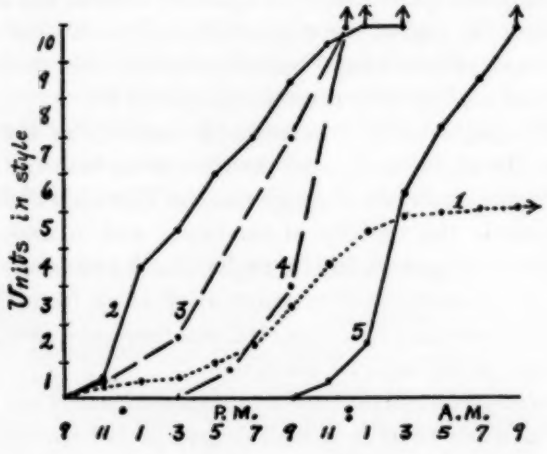


Fig. 1. Curves for pollen-tube growth for the Europa daylily: (1) selfed, normal pollination; (2) ×*H. Thunbergii*, normal pollination; (5) ×*H. Thunbergii*, delayed pollination; (3) ×*H. aurantiaca*, normal pollination; (4) ×*H. aurantiaca*, somewhat delayed.

made to date only 31 capsules have matured and the largest number of seeds having embryos that were obtained in any capsule has been five (Stout, 1926, fig. 4). In this case the failure to yield capsules and seeds more abundantly to inter-specific cross-pollinations is obviously due to the very marked abortion of macrospores and not merely to a lack of affinity in the relations of fertilization or to the failure of pollen tubes to reach the ovary. The low production of seeds after various cross-pollinations is not due fundamentally to selective fertilization, but to macrospore abortion which follows the abnormalities in sporogenesis. There are hence in this clon two types of sterility, one involving abortion of spores and one involving self-incompatibility in the reactions of pollen tubes of microspores that do not abort.

As shown in figure 1, in the Europa daylily the pollen tubes from self-pollinations penetrate only to about half the length of the styles. From the 6th to the 16th hour a slow rate of growth is rather uniform but almost no advance is made thereafter. Pollen tubes are however not abundant at the lowest point of penetration, and the majority of the tubes remain in the region of the stigma. The ends of the tubes are as a rule only slightly swollen and enlarged.

When the pollen of *H. Thunbergii* is used in cross-pollination on the Europa daylily there is a rapid advance of tubes during the 3rd and the 4th hours, then there is a rather steady rate of growth with, however, a slower rate during the two hours before the ovary is entered. Sixteen hours after pollination pollen tubes are found within the cavity of the ovary in ample time for abundant fertilization. Capsules begin to develop to nearly every flower thus pollinated, but seeds rarely mature after such pollinations evidently because of the abortion of macrospores.

When pollen of *H. Thunbergii* was used in delayed pollination, made 12 hours after the usual time of pollination (i.e. at 9:00 P.M. instead of 9:00 A.M.) the pollen tubes grew somewhat more slowly during the first four hours and then continued at a more rapid pace and were within the ovary at the end of 12 hours. But even in this case the most rapid rate of advance was between the 2nd and the 6th units of measurement of the styles and not in the basal half.

The advance of pollen tubes in styles of the Europa daylily after normal pollination and after a delayed pollination of six hours with pollen of *H. aurantiaca* is also shown in text figure 1. There was a rather uniform acceleration in the growth of the more advanced tubes until they were found within the ovary 15 hours after pollination. At this time, however, the majority of the tubes were in the uppermost quarter of the style, others were scattered at different levels below and only a few entered the ovary. This condition also prevailed three hours later. In delayed pollinations of flowers in the same set of flowers made at 3:00 P.M. or six hours later than the normal pollinations the rate of growth was greater, the acceleration was more pronounced, and the more advanced tubes were in the ovary after 9 hours of growth and at the same hour of the day when pollen tubes of normal pollination reached the ovary. No seed has been obtained from any of these cross-pollinations.

In premature pollination with pollen of *H. aurantiaca* made at 9:00 P.M. on the evening before a set of flowers opened and 12 hours before normal pollination, the pollen remained on the stigma but failed to germinate and penetrate into the pistil until almost the hour for normal pollination. Collections of pistils thus prematurely pollinated were not made after 26

hours, but at that hour of the day the pollen tubes were somewhat more advanced than for normal pollination. This result is not represented in figure 1.

Studies were also made of pollen-tube behavior when pistils of the Europa daylily were decapitated at various levels and pollen applied to the exposed end of the part left with the ovary. When only the stigmas were removed for self-pollination there was some germination of pollen and penetration of the pollen tubes but none grew better or extended further than when the stigmas were not removed. When the upper four-fifths of the style was removed and self-pollen applied to the cut surface a

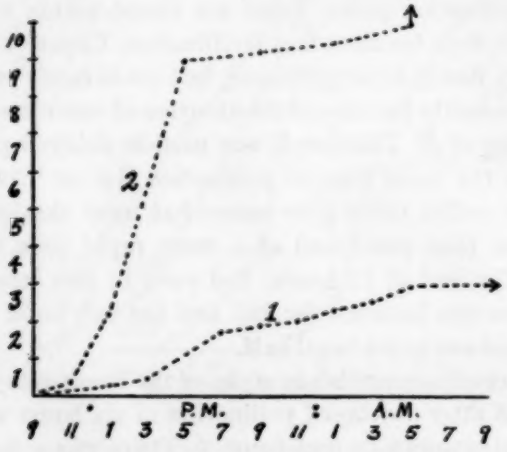


Fig. 2. Curves for pollen-tube growth: (1) of *H. fulva* clon Maculata, selfed, which is completely self-incompatible, and (2) for a wild plant of *H. fulva* selfed, which is somewhat self-compatible.

few pollen tubes grew through the lower part of the style and extended into the ovary but none were found entering a micropyle. Pollen of *Hemerocallis Thunbergii* was also applied to pistils cut to one-fifth their length, but in no case did pistils thus treated give mature capsules with seeds.

*Pollen-tube growth in the H. fulva clon Maculata.* This garden clon has been in cultivation since 1897 and it originated either from a seed or from a living plant sent from Shen-si, China to Florence, Italy. This clon is triploid with $3n = 33$ chromosomes, but it is quite distinct from the older Europa daylily, in having larger flowers and slightly different coloration (for colored plate and history, see Stout, 1929). There is much abortion of pollen and there is complete self-incompatibility, for the pollen tubes which do develop penetrate only a short distance down the stylar canal

(fig. 2). In certain cross-pollinations with other types of fulvous daylilies and especially with all triploids thus far tested the clon Maculata is completely seedless. In cross-pollination with plants of *H. citrina* as many as fifteen seeds have been obtained in a capsule.

*Pollen-tube behavior in a wild plant of H. fulva.* A wild plant, *H. fulva*, obtained directly from the interior of China has been rather highly self-compatible in that numerous capsules with from 1 to 15 seeds each are obtained to selfing. The study of a set of pistils indicates that the pollen tubes from selfing (fig. 2) advance rapidly and at a very uniform rate from 2 to 8 hours when they are almost at the opening of the ovary. Then they

TABLE 1

*Results of successive self-pollinations of one large plant of Hemerocallis Thunbergii*[a]

| DATES OF POLLINATION | JUNE | | JULY | | | | | | | | | | | | | TOTALS |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | 28 | 30 | 1 | 3 | 5 | 7 | 8 | 10 | 11 | 12 | 13 | 14 | 15 | 17 | 18 | |
| No. flowers | 4 | 14 | 11 | 15 | 6 | 14 | 7 | 16 | 19 | 7 | 5 | 11 | 10 | 6 | 8 | 153 |
| No. complete failures | 4 | 11 | 5 | 6 | 6 | 10 | 5 | 12 | 13 | 4 | 2 | 4 | 5 | 3 | 4 | 94 |
| No. rudimentary capsules | | 2 | 2 | 2 | | 1 | 1 | 1 | 1 | 1 | 1 | 2 | 2 | 2 | 1 | 19 |
| No. mature capsules | | 1 | 4 | 7 | | 3 | 1 | 3 | 5 | 2 | 2 | 5 | 3 | 1 | 3 | 40 |
| No. seeds per capsule | | 3 | 1–3 | 1–4 | | 1–4 | 3 | 1–4 | 1–2 | 1–3 | 1 | 1–2 | 1–4 | 1 | 1–2 | 80 |

[a] This plant of *H. Thunbergii* bloomed from June 28 until July 18. A total of 150 flowers were self-pollinated, 91 ovaries soon fell, 19 remained to form dry empty capsules, and 40 gave a total of 80 seeds, the number ranging from 1 to 4 per capsule. This is typical of results thus far obtained with all daylilies that are feebly self-compatible.

linger in this position for about 12 hours before they enter the ovary. This is a striking case of a delay of pollen-tube growth at the opening into the ovary followed by the advance of some tubes into the ovary and some fertilization.

*Pollen-tube growth in Hemerocallis Thunbergii.* The clon of *H. Thunbergii* studied is one that has been in general garden culture since 1890 (Stout, 1929, for colored plate and description). It is diploid ($2n = 22$), the pollen is highly potent, and capsules and seeds are readily produced to certain crosses. After self-pollinations the rule is that the perianths are shed leaving the ovaries which soon begin to enlarge. Many of these ovaries absciss from 7 to 26 days after pollination; others remain attached though they shrivel to small dried structures (Stout, 1921, fig. 8), and about one-third of all flowers pollinated will yield capsules which mature and contain from 1 to 5 seeds each with occasionally as many as eight seeds. Such capsules require from 40 to 55 days to mature. There is no cyclic mid-period or end-period self-compatibility, for the production of these capsules is

distributed throughout the entire period of flowering as is shown in the data in table 1. The maximum number of seed is always very low, quite as is characteristic of many cases of partial compatibility.

After normal self-pollination in the clon of *H. Thunbergii* studied, the rate of pollen-tube growth is very rapid during the 5th, 6th, 7th, and 8th hours at the close of which the ends of tubes are within 1 cm. of the ovary. But they linger in this region for about 8 hours and are first found within the ovary at the end of 18 hours after pollination. There is only slight swelling of the ends of the pollen tubes and there is evidently no

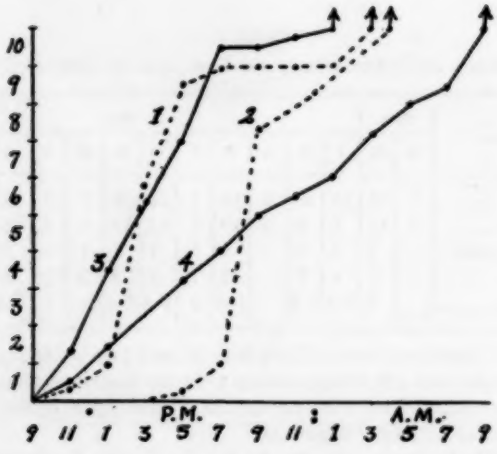


Fig. 3. Curves for pollen-tube growth for *H. Thunbergii:* (1) selfed, normal; (2) selfed, delayed; (3) ×*H. aurantiaca*, normal; (4) ×Europa daylily, normal.

bursting. Here an inhibition in the advance of the tubes takes place near the entrance to the ovary. But tubes finally enter the ovary and at least some fertilizations may result to give the few seeds obtained.

Pollen-tube behavior was studied after delayed self-pollinations made six hours later than normal. For these, the pollen tubes grew slowly during the first four hours, then there was a rapid growth for two hours during which nearly seven-tenths of the style was traversed, and then the growth was decidedly retarded during the 7 to 9 hour period. No collections were made between the 9th hour and the 13th hour at the end of which many tubes were within the ovary. After delayed self-pollination the tubes reach the ovary more quickly than after normal self-pollination due to more rapid growth during the 4–6 hour interval and to less retardation in the region of the ovary.

After inter-specific-pollination with pollen of *H. aurantiaca* the rate

of pollen-tube growth is rapid during the first ten hours at the close of which the ends of the tubes are less than 0.5 cm. from the upper end of the ovary. But the ends of the tubes remain in this position and do not enter the ovary until some six hours later. This cross readily yields capsules and viable seeds. In certain crosses on this clon of *H. Thunbergii* nearly every flower that is pollinated yields capsules filled with seeds, the number of which has been as high as twenty-three, a result which demonstrates that the ovules are highly able to function when there is the proper fertilization.

When the pistils of this plant are pollinated with pollen of the *H. fulva*

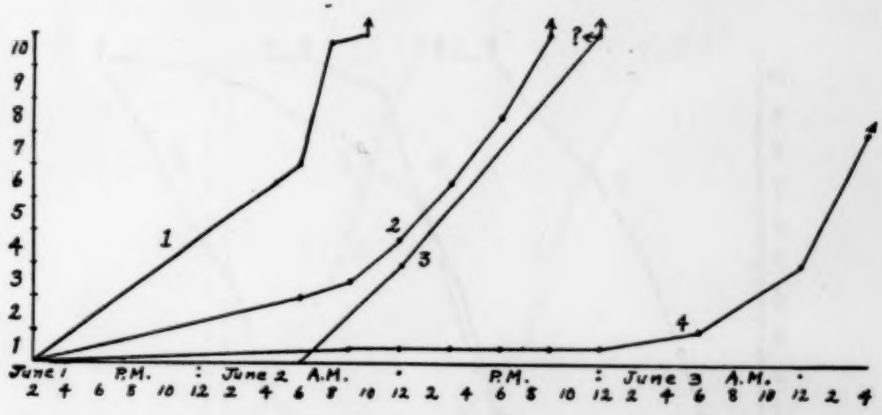


Fig. 4. Curves of pollen-tube growth for *H. flava:* (1) delayed pollination on flowers fully open on June 1; (2) premature pollination of set to fully open on June 2; (3) normal pollination of set fully open on June 2; (4) very premature pollination of set to open on June 3rd.

clon Europa the pollen tubes grow steadily but at a much slower rate for a period of 22 hours when there is an acceleration which brings them into the ovary at the end of 24 hours. But fertilization does not readily occur or at least few capsules with viable seeds mature. Of 1895 flowers of this cross only 8 capsules matured and the number of seeds per capsule ranged from 1 to 5, but thus far no seedlings have been reared from such seed.

*Pollen-tube behavior in Hemerocallis flava.* The clon of *H. flava* which was studied (Stout, 1929) is highly self-fruitful for it produces fine large capsules well filled with viable seeds to almost every flower that is self-pollinated. It is obviously a different clon from that of the *H. flava* studied by Focke (1893) which was reported to be self-fruitless, and also from that studied by Jost (1907) which was feebly self-fruitful. The description of Jost for the self-sterility of the plant he called *H. flava* applies almost

exactly to the results seen in *Hemerocallis Thunbergii*. Some of the seedlings of the clon here studied appear to be self-incompatible, and so there may be many incompatibilities within this species and in the genetical nature of the clon here studied. The particular clon studied is, however, highly self-fruitful as far as the production of seeds to selfing is concerned. Plants of this clon of *H. flava* bloom early in spring. During warm weather the sets of flowers open in the late afternoon and continue in excellent condition throughout the following day. With cooler weather the sets may not start anthesis until in the early forenoon. A single set may remain open and fairly fresh for a period of 50 hours.

Fig. 5. Curves of pollen-tube growth for an $F_1$ hybrid (Ser. 6 #1) of *H. flava* × *H. fulva* clon Europa: (1) selfed, premature; (2) selfed, normal; (3) selfed, delayed; (4) ×a sister hybrid, normal.

In this clon the data from two collections of pistils after a self-pollination at 6:00 A.M. of flowers open for that day indicate that pollen tubes are within the ovary at least at the end of 18 hours. Special studies were made for premature self-pollinations and these showed that while some grains promptly germinate the tubes grow slowly until about the time of the early forenoon when normal pollinations occur. This was very marked in the case of a premature pollination on June 1st of flowers that were open on the day of June 3rd.

*Pollen-tube behavior in the two $F_1$ hybrids of H. flava × H. fulva clon Europa.* Pollen-tube behavior was determined for two of the $F_1$ seedlings (Ser. 6 #1 and Ser. 64 #2) obtained by crossing *H. flava* as a seed parent with the Europa daylily. These seedlings are therefore hybrids between

two distinct species and of the individual plants used as parents one is self-fruitful and the other is self-incompatible.

The hybrid, Ser. 6 #1, is very feebly self-compatible; from many self-pollinations only two seeds have been obtained. As shown in text figure 5, the pollen tubes of normal self-pollinations grow rapidly from 2 to 6 hours after pollination; then their rate is retarded and they grow much more slowly and a few enter the ovary only after 22 hours. A set of flowers was also utilized in delayed self-pollination; flowers were emasculated and bagged, and the pollen from other flowers was saved from forenoon until 9:00 P.M. when the delayed pollinations were made. For these pollinations

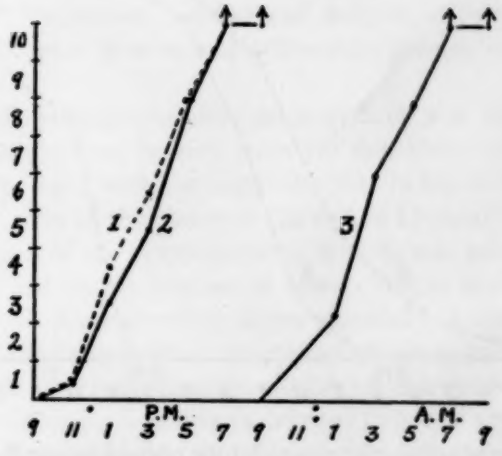


Fig. 6. Curves of pollen-tube growth for a hybrid (Ser. 64 #2) of *H. flava* × *H. fulva* clon Europa: (1) selfed, normal; (2) × *H. Thurnbergii*, normal; (3) × *H. Thunbergii*, delayed.

the tubes continued at a rather rapid and uniform pace and without delay entered the ovary after 10 hours of growth at the same hour (7:00 A.M.) of the day when the tubes of pollinations made ten hours earlier on the same day (at 9:00 A.M.) entered the ovary.

After premature self-pollination the pollen tubes grow at a rather rapid rate after the first two hours and enter the ovary within a period of 14 hours yet no seed was produced to the controls of such pollinations. In this plant the normal pollinations result in much delay of pollen tubes in the region near the ovary, while in premature and in delayed pollination there is no such delay.

When the plant 6 #1, which is diploid, is pollinated with pollen of its triploid pollen parent, Europa, the pollen tubes penetrate only about one-

tenth of the length of the style and hence no fertilizations result. This is a very complete cross-incompatibility with the pollen-tube growth completely inhibited in the apical portion of the style.

The hybrid 64 #2 rarely sets a seed to self-pollination, yet the pollen tubes grow rapidly and steadily beginning with the 3rd hour and at the end of the 10th hour are within the ovary (text figure 6). Here is a most striking case of reaction of self-incompatibility within the ovary. When crossed in normal pollination with *H. Thunbergii* the tubes grow almost as in the selfing and enter the ovary after the same interval of time, but in

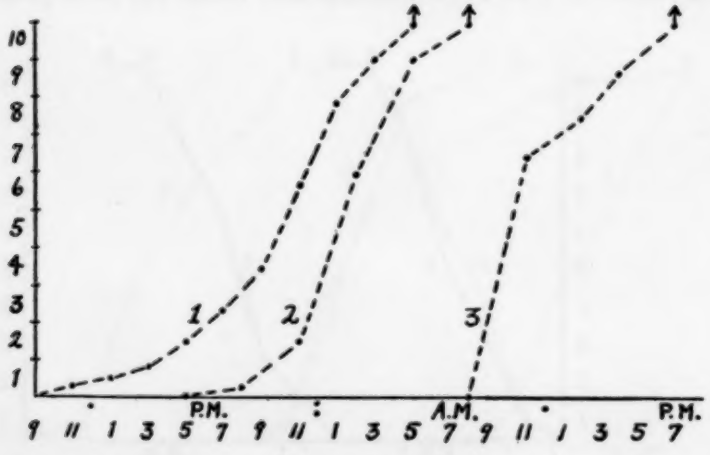


Fig. 7. Curves of pollen-tube growth for the night-blooming *H. citrina:* (1) selfed, premature; (2) selfed, normal; (3) selfed, delayed.

this case there is the development of capsules with numerous viable seeds. The curve for pollen-tube growth after delayed cross-pollination with *H. Thunbergii* is almost identical with that for normal pollination.

Judged by seed production to controlled self-pollination the two sister plants, 6 #1 and 64 #2, are equally self-incompatible but the pollen-tube behavior for normal pollinations is very different; for 6 #1 there is inhibition of growth in the region immediately above the ovary; for 64 #2 there is no delay in the advance of tubes and the incompatible reactions occur within the ovary. But in premature and in delayed self-pollination of 6 #1 the behavior is quite the same as that of 64 #2 for normal pollination.

*Pollen-tube behavior in Hemerocallis citrina.* Several different clons have been received under the name *Hemerocallis citrina* but the one studied was obtained from Willy Müller in Naples from the garden (Hortus Nucerensis) from which this species was first distributed to the trade, and this

clon is said by Mr. Müller to be a division of the stock first described as *H. citrina* (for history, description and colored plate see Stout, 1930). This clon is decidedly nocturnal in flowering habit; the flowers open in the evening and shed pollen during the night, but usually there is abundant pollen present in anthers the next morning. If the day is warm and sunny the perianths wilt early in the forenoon but if the weather is cool and cloudy the flowers continue more or less open and fresh until at least midday. For this clon normal pollinations are considered to be those made in the evening on freshly opened flowers.

The clon of this species which the authors studied is self-incompatible to the extent that very few seeds have thus far been obtained to numerous self-pollinations. Premature, normal and delayed self-pollinations were made on a single set of flowers and pollen-tube growth for each was determined.

The "normal" pollinations were made at 5:00 P.M. when pollen was shedding in freshly opened flowers. After the first three hours pollen-tube growth was fairly rapid with the maximum rate in the middle part of the style, and tubes were in the ovary at the end of 15 hours. For premature pollinations, pollen of the previous set of flowers was saved in vials and applied at 9:00 A.M. to the stigmas of flowers still in bud. For these the pollen-tube growth was somewhat slower especially in the apical portion of the pistil and the tubes entered the ovary only three hours earlier in the day than they did after normal pollination. After delayed pollination made at 8:00 A.M. the following forenoon (a delay of 15 hours) the rate of growth of tubes was very rapid for the first three hours during which three-fifths of the style was traversed. Then the rate became much slower and the tubes were in the ovary at the end of 12 hours.

It is to be noted that certain inter-species cross-pollinations on this clon of the *H. citrina* yield seeds whether pollinations are made in the evening or in the following forenoon, but that self-pollinations rarely yield seed although the tubes to normal self-pollination reach the ovary at 8:00 A.M. This result seems to indicate that the self-incompatibility in this case involves reactions within the ovary.

## SPECIAL TESTS OF POLLEN-TUBE GROWTH IN CULTURES

The viable pollen grains of daylilies readily germinate on media of 5, 10, or 15 grams cane sugar, 1 gram agar agar, and 100 cc. water at room temperature. Somewhat extended studies were made of the rate and the extent of growth on media for pollen collected at different ages of the flower and at different temperatures for reactions to secretions of the pistil.

The growth of pollen tubes on artificial media and under various constant temperatures was studied for pollen of *H. citrina, H. Thunbergii* and *H. fulva longituba*. The best growth was obtained under constant temperatures ranging from 68° to 80° F., and tubes 1210$\mu$ long were obtained. In all cases the rate of growth was most rapid during the first two hours and in most cases there was little or no increase in length after this period. It is to be noted that the maximum length of growth of pollen tubes on artificial media is in this case only about 1 mm., which in the pistils would scarcely more than carry tubes to the opening of the stylar canal.

Special tests for evidence bearing on chemotactic reactions were made. Pistils were placed on agar media with the stigmatic end near the center of the culture and pollen grains were scattered evenly about the stigma. Self-pollen and cross-pollen of various combinations were tested for various daylilies. In many cases there was no noticeable influence on the direction of the growth of pollen tubes. In several cases tubes in the immediate region of the stigma grew toward the stigma but this was the case for self-incompatibility as well as for cross-fertility. Sections of the style and the ovary were used in the same way as the stigma but with no definite results. Also the fluids obtained from severed styles and ovaries were drawn into fine capillary glass tubes which were placed in the midst of cultures or with one end resting near the periphery of a culture. There was excellent germination and growth of tubes but in no case was there any recognizable effect on the growth of tubes either in the length to which tubes grew or in the direction of their growth. Droplets of the fluid obtained when styles are severed were placed on slides and pollen grains were sown directly in them but the grains burst and no germination was obtained which would seem to indicate that the stylar canal of *Hemerocallis* does not contain the same fluid that is obtained when the style is severed and which evidently comes largely from the cortex.

## SUMMARY AND DISCUSSION

The main aspects of pollen-tube behavior most characteristic for the compatible fertilizations after normal pollination in *Hemerocallis* may be stated as follows:—the pollen tubes travel in an open canal throughout the entire length of the pistil; a period of about two hours after pollination is usually required before tubes are noticeably within the stylar canal; the most rapid rate of pollen-tube advance is in the upper-middle portion of the style; fertilization is completed within a period of 24 hours after normal pollination.

Obviously, characters of generic specificity are responsible for many

aspects of pollen-tube behavior and for marked differences in the behavior among different genera of flowering plants. Thus for comparison, in highly compatible cross-fertilizations between plants of *Nicotiana Forgetiana* at least five days are required (East and Park, 1918, fig. 1, p. 361) for the pollen tubes to traverse styles that are only 2.0 to 2.5 cm. in length. The relatively much more rapid advance of pollen tubes in *Hemerocallis* is correlated with the longer styles, the shorter life of pistils, and possibly also with the hollow rather than the solid style. The long life of the pistils of *Nicotiana* allows pollen tubes of self-incompatible relations to continue growing for as many as 14 days (East and Park, 1918, fig. 1), a condition not possible in *Hemerocallis*.

It is to be noted that there are numerous types of selective and discriminative relations in fertilization. Inter-specific and inter-varietal relations, the relations between various polyploids, the various interactions of intra-specific incompatibilities, and the various grades of self-incompatibility are all expressed in the reactions in the pistil. The scope for pollen-tube reactions is so limited that distinctive and specific behavior can not be expected for each of the various sorts of selective action. The reactions observed may however be considered and properly related to the particular condition known to exist from experimental data.

In the various forms of *Hemerocallis* studied three distinct types of pollen-tube behavior in self-incompatibility are to be recognized:

(1). The reactions may be complete in the upper portion of the style. In this case the tubes travel slowly and only advance to a short distance in the stylar canal. This was seen only after the selfing of the two triploid clons Europa and Maculata. The type of reaction which is expressed in the stigmatic end of the pistil has been known in the earlier studies of Jost (1907) and of Correns (1913) and in the more recent studies, especially of *Nicotiana* by East and Park (1918). Such relations in solid pistils, it would seem, involve most completely a relation between haploid pollen tubes and diploid tissues of the style. In the triploid daylilies it may be an expression of a doubling through triploidy of certain of the hereditary factors for incompatibility.

(2). In many cases of self-incompatibility in *Hemerocallis* the pollen tubes are at or near the open entrance to the ovary in ample time for fertilization but there is then a decided inhibition in the advance of the tubes. In such cases it appears that the tubes are reacting to secretions from the ovary. In certain cases the tubes may linger or remain almost stationary for several hours and then proceed into the ovary. Possibly this reaction indicates that special inhibiting substances are no longer being produced and that any such substance produced earlier becomes in-

active. During the time when the advance of a group of pollen tubes is checked either temporarily or permanently the shorter tubes behind them do not continue to advance.

The pollen tubes in *Hemerocallis* have been examined rather carefully for any evidence of differential growth among classes of sister pollen. The pollen tubes are more or less scattered but convincing evidence of two or more different modes or levels in their distribution has not been observed. Such reactions are assumed to occur for certain cross-pollinations between plants which carry factors for incompatibility. Thus in the simpler type of incompatibility crosses between two plants which possess one factor in common (as $S_1S_2 \times S_1S_3$) only one class of pollen (the $S_3$) is assumed to function (see especially Lehman, 1927). Also in the type which involves qualitative and quantitative inter-relations between incompatibility factors and factors for fertility as conceived by Kakizaki (1930) differential fertilizations in certain crosses are assumed to occur. Thus in the cross $S_1S_2T_2T_2 \times S_1S_1T_1T_2$ only the $S_1T_2$ pollen is supposed to function. But in neither of these types of incompatibility is selective fertilization assumed to occur in selfing. Selective fertilization in selfing is, however, well known for various plants in which certain pollen-tube factors are linked with endosperm qualities that show deficiency in ratios as seen in corn (summarized by Jones, 1928). Direct observation of bimodal distribution of pollen tubes from sister pollen has been made in the case of crosses between $2n \times 2n+1$ plants of *Datura* (Buchholz and Blakeslee, 1932) and it is considered that the same differential growth occurs in selfing $2n+1$ plants. In the types of *Hemerocallis* studied differential fertilizations in selfing are not apparent in pollen-tube behavior.

(3). There are cases of almost complete self-incompatibility in which the pollen tubes proceed directly into the ovary without delay, and yet seeds are seldom formed. In this condition it is clear that the reactions of incompatibility involve either (a) the pollen tubes and the secretions of the ovary or possibly of the ovules or of the egg apparatus, or (b) the relations of sperm and egg in the final stages of fertilization, or (c) the abortion of young embryos.

When the reactions are close to or within the ovary there is obviously greater opportunity for a direct relation between haploid pollen tubes and the haploid macrogametophytes, and especially when there is a stylar canal.

Reactions of incompatibility after the pollen tubes reach the ovary or enter it are to be recognized as a well-defined type of behavior. This condition has been noted in certain other plants which exhibit self-incompatibility, namely, in certain apples (Cooper, 1929) and for certain individuals

of head cabbage (Sasaoka, 1928). In the cyclic self-incompatibility of plants of *Brassica pekinensis* (Stout, 1931*b*) powerful reactions may operate in the first flowers to open which produce coiling of pollen tubes on the stigma but in certain later flowers the tubes may enter the ovary and yet no seeds, or only a very few seeds, result.

In a series of studies with *Petunia*, Yasuda, (1932) concludes that the substances which inhibit pollen-tube growth in the secretions of the stigma are mainly secreted in the ovary and then they move to the upper part of the stigma.

In the various cross-fertilizations studied there were no cases of inhibition of pollen in the upper portion of the style. In the cross *H. Thunbergii*×*H. aurantiaca* (fig. 3) the pollen tubes were delayed for several hours at the entrance of the ovary quite as in the selfing. For the crosses of the Europa daylily with pollen of *H. aurantiaca* and *H. Thunbergii* the pollen tubes proceed without delay into the ovary. Thus the pollen-tube behavior in the cross-relations studied shows no noticeable inhibition in the upper portion of the style. In these inter-species fertilizations factors of specificity rather than those of incompatibility are no doubt involved.

A low yield of seed, or "feeble compatibility," to certain selfings and crossings is characteristic of incompatibilities not only in daylilies but in numerous other plants. This is evident when a particular pollination which is amply and properly made very constantly gives less seeds than are possible when the seed parent and the pollen involved are used in other relations. In the daylilies this condition involves, as a rule, reactions at or within the ovary rather than merely a slow rate of growth of pollen tubes in the style. Also seeds are not increased in number by premature pollination. The studies show rather definitely that in many such cases pollen tubes reach the ovary at the time when tubes are normally attracted to the egg apparatus and when a full complement of seeds is possible. Under such conditions the low seed number can be due either to the presence in the ovary of few tubes or to reaction of incompatibility and selective fertilization in the ovules.

The behavior of pollen tubes in *Hemerocallis* makes it clear that the reactions of incompatibility may be complete in the upper part of the style as seen in a few cases, but that it is the rule that they are expressed at the entrance to the ovary or within the ovary.

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A preliminary study of pollen-tube growth in several types of *Hemerocallis* was made during 1927 by Mr. T. Susa while technical assistant at the New York Botanical Garden. It was deemed advisable to extend the

studies to include other types and relations, and to make more frequent collections of a larger number of pistils in each relation and for each pollination, and this was done for the data here reported.

NEW YORK BOTANICAL GARDEN

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**Explanation of plate 23**

Fig. 1. Pistils of *Hemerocallis Thunbergii* (A) and of Europa daylily (B). About natural size.

Fig. 2. Stigmatic portion of pistil showing upright papillae soon after flower opens. ×18.

Fig. 3. Cross section of pistil of *H. Thunbergii*, at *a* of fig. 1. ×35.

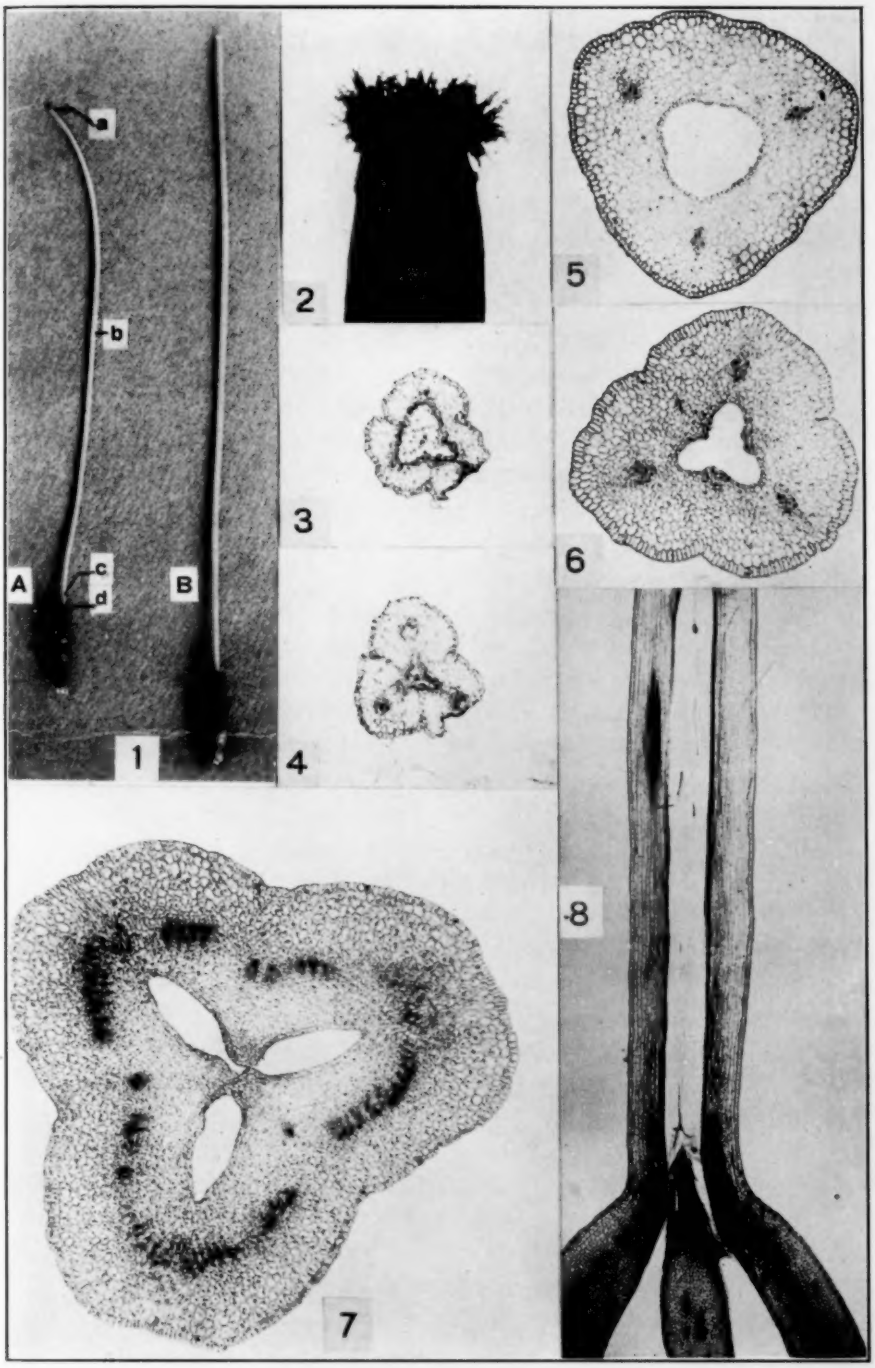
Fig. 4. Cross section of pistil just below *a* in fig. 1, at point where stylar canal is constricted. ×35.

Fig. 5. Cross section of pistil at *b* of fig. 1, showing large stylar canal. ×35.

Fig. 6. Cross section of pistil at *c* of fig. 1, showing entrance to the three ovary chambers. ×35.

Fig. 7. Cross section of pistil at *d* of fig. 1, showing the beginning of the ovary. ×35.

Fig. 8. Longitudinal section of *H. Thunbergii* pistil showing pollen tubes in the style 24 hours after self-pollination. ×21.




STOUT AND CHANDLER: HEMEROCALLIS

UNIV. OF MICH.

## Preparation of pollen for microscopic examination

R. P. Wodehouse

The question has often arisen regarding whether a wet or a dry medium is to be preferred for mounting pollen, and upon which may be placed the greater reliance—this because most kinds of pollen are extremely sensitive to changes in moisture owing to certain hygroscopic substances which they contain; and frequently there is much difference between their appearance when wet and when dry. Most kinds of pollen, except those with very thin, elastic or fragmentary exines, have their own peculiar ways of accommodating changes in volume with corresponding changes in form. Indeed, the mechanisms by which this is accomplished are enormously varied and sometimes elaborate and extremely complicated, and they show that volume-change-accommodation is a real and normal function of the exines of pollen grains.[1] And as we pass in review the pollen of any considerable number of different species of plants it becomes quite evident that the mechanisms for the accomplishment of volume-change accommodation have undergone evolutionary development comparable in extent to that of the adaptive mechanisms of flowers to pollination by insects. In all studies of pollen-grain morphology, therefore, mounting media which reveal such mechanisms must be chosen.

Most frequently volume changes are accommodated by three or more slit-like furrows in the exine. When there are only three furrows they are generally meridionally arranged, but when there are more, they may have other arrangements.[2] When such a grain is dry its furrows are generally closed and so far buckled inwards that not even their margins may be seen, but as soon as it is moistened and expanded its furrows open widely; then it is seen that each is crossed by a delicate furrow membrane which may or may not possess decorations of its own, but is usually marked at its center by a germinal aperture of varying shape, and through which a germinal papilla bulges, and occasionally each furrow is crossed by an underlying transverse furrow. Since the opening of the furrow, therefore, reveals the furrow margins, the germinal aperture and papilla, and the transverse furrow when present, a grain that is expanded is enormously more interesting and instructive than one that is dried and collapsed, and in which none of these things can be seen.

It should be stated at this juncture that this view is by no means universally accepted. All of the earlier investigators believed that pollen could be satisfactorily observed only in its "dry and natural" form, in

[1] Wodehouse, R. P. Ann. Bot. **42**: 896. 1928.

[2] Wodehouse, R. P. Bull. Torrey Club **57**: 21–46 1930.

ignorance of the fact that the wet and the dry forms are equally natural. The impression of the unnaturalness of the forms assumed by pollen grains when wet was corrected by von Mohl[3] in 1835 and he also showed that much more could be seen in them in their moist than in their dry condition. Very recently, however, my work has been severely criticized by botanists of eminence in their own field for having been done with grains in their expanded form. Ferguson and Coolidge,[4] in their studies upon petunia pollen, state: "In his studies of pollen grains, Wodehouse (1928) used as mounting media either an alcohol-water-glycerine combination or Brandt's glycerine jelly. . . . From the data presented in the present paper it seems clear that both dry air and balsam as mounting media give a truer picture of the size and form of pollen grains than does any aqueous medium." And again: "As a result of our observations it is believed that descriptions of pollen grains, as observed in an aqueous medium are of doubtful value." It is consoling that, as the subject of this criticism, I have the company of such eminent investigators as Jeffrey, East, Blakeslee, Sax, Brink and others. For, in referring to the pollen work of this list of investigators, and with which my own name is included, they state: "Grains were first studied and measured in the various media used by other writers: water, sugar solutions of different degrees of density, fuchsin, lactic acid. . . . The results with the several media were in general the same; but in all of them one is studying and comparing *transformed* pollen grains, not the true normally shaped grains."

When a dry pollen grain is moistened and expanded, admittedly it changes its shape. But very often it is in this condition when it leaves its anther. Ferguson and Coolidge admit that this is so even with petunia pollen on a rainy day, but after a short exposure to dry air it loses its turgidity and presumably becomes more normal. Beyond question such changes in volume and shape must always be ascertained, and the investigator must remain cognizant of them in making deductions, whichever condition he chooses to regard as normal. If the grain has three furrows, which is the commonest number, it generally changes upon being moistened, from ellipsoidal or cylindrical in form to globular or even oblately flattened. But with these changes a further *transformation* takes place. The furrows open. The opening of a pollen-grain furrow may be likened to the opening of the human eye; the furrow margins correspond to the lids, the furrow membrane to the whitish sclerotic coat, the germinal aperture with its papilla to the iris with its pupil. Just as it is more interesting and instructive to observe an eye that is open than one that is

[3] Mohl, H. von. Ann. Sci. Nat. Ser. 2. 3: 148. 1835.

[4] Ferguson, M. C., & Coolidge, E. B. Am. Jour. Bot. 19: 644. 1932.

closed, so it is more interesting and instructive to observe a pollen grain that is expanded with its furrows open than one that is collapsed with them closed.

## METHODS

*Methyl-green-glycerine-jelly.* A small amount of pollen, about as much as can be picked up on the flat end of a tooth pick, or less, is placed on the center of a microscope slide, a drop of alcohol added and allowed to partly evaporate. A second and third or even fourth drop may be added if necessary. The alcohol spreads out as it evaporates and leaves the oily and resinous substances of the pollen deposited in a ring around the specimen. The oily ring is wiped off with cotton moistened with alcohol, and, before the specimen has had time to completely dry, a small drop of hot melted methyl-green-glycerine-jelly is added and the pollen stirred in with a needle and evenly distributed. During the process the jelly is kept hot by passing the slide over a small flame, heating it just enough to sting but not burn the knuckle which may be used to test its temperature. A number 0 cover glass, which has been passed several times through the flame while held vertically with the forceps, is then placed over the specimen, and the slide gently heated. If the amount of jelly has been judged correctly the cover will settle into position with the gelatin reaching its periphery just when the pressure of the cover begins to be taken up by the pollen grains. This amount must be learned by experience and accurately gauged because a smaller amount leaves the grains crushed or flattened by the cover, or the mount incompletely filled, and a larger amount causes the preparation to be too thick for use with oil-immersion lenses.

If naturally shed pollen is not available, satisfactory material can generally be obtained from herbarium specimens, providing they were quickly and completely dried. Often it is only necessary to tap the dry flowers over the slide or crush a few anthers on it. If pollen cannot be removed in this way, a few anthers, or with the Compositae, a few florets may be removed from the specimen and placed on the slide. These are then moistened with alcohol, followed by a drop of water and heated to boiling. The pollen may then be teased out and the anthers and other debris removed, leaving the pollen in the water. The water is then drawn off with cotton or filter paper and the jelly added as before.

The glycerine jelly is prepared according to the method of Brandt (Lee, Microtomist's Vade Mecum, 7th Ed. p. 242), which is as follows. Soak some gelatin for two or three hours in cold water, pour off the superfluous water and heat until melted. To one part of this add one and one half parts of glycerine and, while still hot, filter through spun

glass pressed into the lower part of a heated funnel. Add two or three per cent phenol. Still keeping the mixture hot and fluid add, drop by drop, a saturated solution of methyl green in fifty per cent alcohol, until the glycerine jelly becomes fully as dark as green ink.

The relative proportions of glycerine and water are so balanced in this medium that the majority of pollen grains when placed in it are fully but rarely over expanded, and they never do burst and extrude their contents as is usually the case with ordinary aqueous media.

### ATMOSPHERIC POLLEN SLIDES

The same medium is the best, in my experience, for catching atmospheric pollen and making pollen counts and identifications. To do this, a small drop of melted methyl-green-glycerine jelly is placed on a slide, spread out to occupy an area about equal to and of the same shape as the cover glass which is to be used in finishing the mount. The slide is exposed in a horizontal position protected from rain and sun by a shelter raised at least four inches above it. After twenty-four or forty-eight hours the slide is brought into the laboratory and examined with a hand lens, and any extraneous material like soot or sand which, if present, might prevent the cover glass from fitting into place, is removed. The slide is then heated, controlling the temperature with the knuckle as before, until any excess moisture which may have accumulated during the exposure, is driven off. It is then covered with a number 0 cover glass.

### EXAMINATION OF DRY POLLEN

When it is desired to discover the unexpanded shapes of pollen, it may be observed in dry air with or without a cover glass. Such observations are also useful in determining the amount of oil naturally occurring on the surface of the grains, and as a check against the method given below; but under these conditions magnifications over two hundred times can scarcely be used to advantage. Nevertheless, even at this magnification the shapes of the grains can be learned and something of the mechanical action of their organs of volume-change-accommodation. If, however, a detailed examination of the grains in their unexpanded condition is desired, they must be stained and brought into a medium of suitable refractive index. This may be done as follows:

### ANALINE OIL GENTIAN VIOLET METHOD

Place the pollen on the slide as before and add two or three drops of analine oil which has been tinted with gentian violet only to a pale purple color. Heat gently over the flame, controlling the temperature with the

knuckle as before, until the grains become deeply stained. Allow the slide to cool to room temperature, draw off the excess oil with filter paper, wash by repeatedly adding xylol and drawing it off until all the oil and unabsorbed dye have been removed; then add a drop of Canada balsam and cover. This presents the grains unexpanded but brilliantly stained, and in a medium eminently suited for observation with high-power oil immersion lenses.

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# The growth of Rhododendron ponticum in sand cultures[1]

E. L. Spencer and J. W. Shive

In a study of the mineral nutrition of a plant, there are many factors which must be considered, for the intake of the nutritive elements involves complex chemical and physical processes. Much is already known concerning the nutrition of some plants, such as wheat, soybean, alfalfa and other agricultural crops, under definite experimental conditions. From a review of the literature on the subject of mineral nutrition, it is at once apparent that there is still a great deal to be learned regarding the response of other groups of plants to the mineral elements which are essential to growth.

The large group of plants usually classified as "ornamentals" serves as a typical illustration of the great number and diversity of the species to which little attention has been given from the standpoint of a systematic study of the mineral requirements of plants of such great economic value. It has been only within recent years that this large class has been considered as a field for the scientific investigation of problems in plant nutrition, and this attention has been forced largely by the rapidly growing commercial importance of this vast group of plants.

The data presented in the following pages deal with an important member of this large group. They are the result of an experimental study to determine the mineral requirements for good growth and development, from seed, of *Rhododendron ponticum L.* under experimental conditions carefully controlled, in so far as this is possible, with respect to the media in which the plants were grown.

It is recognized that it is not possible to determine the nutrient requirements of a plant growing in the soil, because of the complexity of such a medium which cannot be controlled or varied at will with any degree of satisfaction or accuracy, owing to the numerous unknown chemical and physical factors involved. Because of this handicap, sand was used as the substratum to which the elements required for growth were added in solution form. Solution cultures serve as the fundamental starting point in the scientific investigation of the mineral nutrition of any group of plants. Furthermore, by means of some adequate system of continuous flow at a constant rate, such as has been developed by Trelease and Livingston (1922), by Anderssen (1926), by Johnston (1927), or by Shive and Stahl (1927), the mineral constituents of a culture solution bathing the roots of an actively growing plant, while they cannot be held at an absolutely constant level, can be maintained indefinitely without any very pro-

[1] Journal Series paper of the New Jersey Agricultural Experiment Station, Department of Plant Physiology.

nounced variations from the original salt or ionic proportions due to the absorption activities of the plant.

The studies here reported mark the beginning of a series of investigations dealing with problems concerned in the nutrition of *Rhododendron ponticum*, a member of the family Ericaceae. They are the response to many inquiries for fundamental information concerning the nutrition and growth of this splendid ornamental which, it appears, is difficult to grow successfully when used as a subject in landscape gardening under conditions favorable for the growth of ornamentals of other families. It was primarily the purpose of this investigation to determine whether culture solutions, such as are known to produce excellent growth of plants belonging to the general class of non-woody agricultural species, would produce similar responses in this woody perennial. Also it was proposed to investigate the properties and possibilities of certain salt combinations in the development of this species, and to make a comparative study of the plants grown by the solution culture method with those produced by the usual commercial practices.

## EXPERIMENTAL MATERIALS AND METHODS

*The culture solutions.* In the preparation of culture solutions the salts used were Merck's Blue Label reagents and Eimer and Amend's chemicals of tested purity. In the preparation of stock solutions the following salts were regularly used: mono-potassium phosphate, magnesium sulphate with its usual seven molecules of water of hydration, ammonium sulphate, and calcium nitrate with four molecules of water. The original stock solutions were made up in large quantities and stored separately in five-gallon carboys. These solutions were made up in 0.5 molar concentration. The distilled water used in this work was produced by a "Barnstead" still and stored in five-gallon carboys similar to those used for the stock solutions The culture solutions were prepared from these concentrated stocks.

In order to vary the salt proportions of the culture solutions by definite increments and still hold the total osmotic concentration of the solutions at a constant level, recourse was taken to the plan adopted by Tottingham (1914). The twenty culture solutions here employed were chosen from the complete series of eighty-four solutions used by Tottingham, with the modification as proposed by Jones and Shive (1921), of substituting ammonium sulphate for potassium nitrate in equivalent partial osmotic concentration. Thus the solutions contained nitrogen in the two forms $NH_4$ and $NO_3$. This series of 4-salt solutions was chosen for investigation with the rhododendron on the basis of previous work by Stahl and Shive (1933), Naftel (1931), Jones and Shive (1921), and Tiedjens and

Robbins (1931) who have shown that $NH_4$-nitrogen when present in the growth medium plays a very important part in the early activities and development of certain agricultural species, and also that, in general, a growth medium containing nitrogen in the two forms in the proper proportions is more efficient than one containing either of these forms alone, other things being equal.

For ease of reference, table 1 gives the solution numbers (column two) of the twenty representative solutions selected from the Tottingham series

TABLE 1

*Partial volume-molecular concentrations of mono-potassium phosphate, calcium nitrate, magnesium sulphate and ammonium sulphate in the solutions employed*

| CULTURE NO. | SOLUTION NUMBER | PARTIAL VOLUME-MOLECULAR CONCENTRATIONS[a] | | | |
|---|---|---|---|---|---|
| | | $KH_2PO_4$ | $Ca(NO_3)_2$ | $MgSO_4$ | $(NH_4)_2SO_4$ |
| 1 | $T_1$ $R_1$ $C_1$ | 0.00211 | 0.00146 | 0.01659 | 0.0014 |
| 2 | $C_3$ | 0.00211 | 0.00438 | 0.01185 | 0.0014 |
| 3 | $C_5$ | 0.00211 | 0.00730 | 0.00711 | 0.0014 |
| 4 | $C_7$ | 0.00211 | 0.01022 | 0.00237 | 0.0014 |
| 5 | $R_3$ $C_1$ | 0.00211 | 0.00146 | 0.01185 | 0.0042 |
| 6 | $C_3$ | 0.00211 | 0.00438 | 0.00711 | 0.0042 |
| 7 | $C_5$ | 0.00211 | 0.00730 | 0.00237 | 0.0042 |
| 8 | $R_5$ $C_1$ | 0.00211 | 0.00146 | 0.00711 | 0.0070 |
| 9 | $C_3$ | 0.00211 | 0.00438 | 0.00237 | 0.0070 |
| 10 | $R_7$ $C_1$ | 0.00211 | 0.00146 | 0.00237 | 0.0098 |
| 11 | $T_3$ $R_1$ $C_1$ | 0.00633 | 0.00146 | 0.01185 | 0.0014 |
| 12 | $C_3$ | 0.00633 | 0.00438 | 0.00711 | 0.0014 |
| 13 | $C_5$ | 0.00633 | 0.00730 | 0.00237 | 0.0014 |
| 14 | $R_3$ $C_1$ | 0.00633 | 0.00146 | 0.00711 | 0.0042 |
| 15 | $C_3$ | 0.00633 | 0.00438 | 0.00237 | 0.0042 |
| 16 | $R_5$ $C_1$ | 0.00633 | 0.00146 | 0.00237 | 0.0070 |
| 17 | $T_5$ $R_1$ $C_1$ | 0.01055 | 0.00146 | 0.00711 | 0.0014 |
| 18 | $C_3$ | 0.01055 | 0.00438 | 0.00237 | 0.0014 |
| 19 | $R_3$ $C_1$ | 0.01055 | 0.00146 | 0.00237 | 0.0042 |
| 20 | $T_7$ $R_1$ $C_1$ | 0.01477 | 0.00146 | 0.00237 | 0.0014 |

[a] Osmotic concentration of each solution = 1 atmosphere.

of 84. These numbers refer to the position which the solutions and cultures occupy in the four-coordinate scheme employed by Tottingham to represent the series in diagram. This table also shows the partial volume-molecular concentrations (gram-molecules per liter of solution) of the salts as they were used. The total osmotic concentration of each solution was calculated to give approximately one atmosphere of possible osmotic pressure. Ferrous sulphate as the source of iron for the plants was added to the culture solutions in aqueous solution from stock freshly prepared each time just before being used. Boron as boric acid and manganese as manganous sulphate were added in concentration equivalent to 0.5 p.p.m. of each to every culture solution.

*The sand and culture vessels.* The white quartz sand which was used

as the substratum with the rhododendron cultures was prepared by washing with a heavy stream of tap water introduced at the bottom of a large container and then allowed to filter up through the sand, the surplus water flowing off over the top of the container. This washing was continued until the overflow water appeared perfectly clear and free of colloidal matter. The surplus tap water was drained off and the sand was then flushed several times with distilled water to remove the tap water retained by the sand.

Three-gallon glazed stoneware percolators were used as culture vessels. These vessels have a sloping, curved bottom with a 2.5 cm. hole in the center. A small watch glass placed over the hole prevents the sand from escaping but still permits free drainage. The vessels were filled with the washed sand to within 1 cm. from the top, and were then ready to receive the plants.

*The plants and their treatment.* The species chosen for this investigation, *Rhododendron ponticum*, is a native of Spain, Portugal and Asia Minor, and is not entirely hardy in this region. Nevertheless, it is very extensively used by nurserymen as an understock upon which most of the common hybrid varieties of rhododendron are grafted. This preference for *Rhododendron ponticum* as an understock is due, undoubtedly, to the fact that the plant is capable of developing an extensive and very efficient root system which reflects its vigor in the development of the hybrid grafts.

The plants used in these investigations were grown from seed in flats 50 cm. long, 35 cm. wide and 8 cm. deep, in a mixture of approximately equal proportions of leaf-mold, garden soil and sand. Because of the long time required for germination and the exceedingly slow growth of the young seedlings, it required from nine to ten months to produce plants large enough for convenient handling. These plants when transferred from the flats to the sand cultures for experimental treatment were about 2 cm. tall.

It cannot here be definitely stated what may have been the influence of this long period of development in the leaf-mold-soil-sand mixture upon the subsequent growth of the plants in the experimental cultures; therefore, in presenting the data this residual influence, whatever it may have been, will be disregarded, since it may reasonably be assumed that all the plants were similarly affected.

In order to obtain some quantitative data on this point, a comparative study was made of seedlings produced as above described and those grown from seed in pure quartz sand to a size sufficiently large for convenient handling in experimental cultures. For this purpose, seed obtained from Holland was sown on the surface of washed quartz sand in flats like

those containing the leaf-mold-soil-sand mixture. The larger grains of sand had previously been removed by sifting through a fine-meshed mosquito netting. During germination the flats were covered with glass to prevent the surface layer of sand from drying out. The flats were kept in a partially covered moist chamber at an average temperature of 16°–18°C. After germination, the seedlings were supplied with a culture solution having a total osmotic concentration of 0.1 atmosphere and the following composition:

$KH_2PO_4$, .00105 m.; $Ca(NO_3)_2$, .000146 m.; $MgSO_4$, .00071 m.; and $(NH_4)_2SO_4$, .00014 m.

This solution has the same proportions of the four salts as solution $T_5R_1C_1$ given in table 1, but is only one-tenth as concentrated as that solution.

TABLE 2

*Comparison of plants grown from seed germinated in a leaf-mold-soil-sand mixture with those from seed germinated in quartz sand*

| SEEDS GERMINATED IN | SEEDLINGS AT TIME OF TRANSPLANTING | | PLANTS AT THE END OF THE EXPERIMENT | | |
|---|---|---|---|---|---|
| | AGE AT TIME OF TRANSFER TO SAND CULTURES (MONTHS) | AVERAGE HEIGHT (CM.) | AGE (MONTHS) | PERIOD OF GROWTH IN SAND CULTURES (MONTHS) | AVERAGE HEIGHT (CM.) |
| Leaf-mold-soil-sand mixture | 9 | 2 | 16 | 7 | 30 |
| Sand | 5 | 3 | 8 | 3 | 33 |

200 cc. of the solution was applied twice daily to the surface of the sand in the flats by means of a small hand sprayer.

The seedlings thus grown in sand and those grown in the leaf-mold-soil-sand mixture, when sufficiently large, were transplanted to the sand in the 3-gallon percolators and were grown for several months by precisely the same methods as were the plants in the three experimental series here to be described. The comparative data of these plants are given in table 2, which is self explanatory. The data show that the plants from seed germinated in the leaf-mold-soil-sand mixture and then grown for seven months by the sand culture method were not as large nor as vigorous at the age of sixteen months as were the plants from seed germinated in quartz sand, at the age of eight months, having been grown in sand cultures by the same methods for only three months. It is important to point out also that the seedlings developed in the leaf-mold-soil-sand mixture were subject to certain diseases which were totally absent from the seedlings developed in the quartz sand.

Three complete series of cultures were carried out in the greenhouse. These will be designated Series A, Series B, and Series C. Each series con-

sisted of 20 cultures, each culture of Series A and C having three plants per culture, and Series B having two plants per culture. Only one series was conducted during any one time period. Series A was carried out from November 1928 to May, 1929; Series B from June, 1930 to February, 1931; and Series C from June 1931 to January, 1932.

When the seedlings were transferred from the leaf-mold-soil-sand mixture to the sand in the 3-gallon percolators, the roots were carefully washed to remove adhering soil particles. The seedlings were then planted in the sand contained in the percolators at equal distances from the center and from each other. The seedlings were selected from large numbers, and care was exercised to choose only those which were as nearly alike as could be determined by observation, in size, vigor, and general appearance.

In order to avoid pronounced changes either in salt concentration or salt proportions of the culture solutions applied to the sand, the solutions were continuously renewed at constant rates by the method of continuous flow described by Shive and Stahl (1927). The continuous flow was maintained at a rate of approximately one liter of solution every 24 hours for each culture. The solution was allowed to drip on a watch glass placed on the surface of the sand in the center of the pot. It then percolated through the sand and dripped from the hole in the bottom of the pot.

Once each week the cultures were thoroughly flushed with distilled water, the purpose of which was to remove any accumulation of salts which might have formed at the surface of the sand due to evaporation. The cultures were allowed to drain for several minutes and the solution renewal by continuous flow was started immediately. Once each week also, the surface layers of sand in each pot were thoroughly loosened by stirring to a depth of one to two centimeters with a pointed glass rod. This prevented excessive evaporation from the surface, aided aeration of the cultures, and practically inhibited the growth of green algae on the surface of the sand.

During the growth period of each series, observations were made of the general character of the development of the plants, and any unusual phenomena or pathological conditions were noted. At the termination of each series, the plants were cut at the surface of the sand. Measurements were made of the height of the stem above the sand and the diameter of the stems at the base. The leaves were separated from the stems and blueprints were made of all the leaves of each plant. From these prints the total leaf area of each plant was obtained by tracing the leaf outline with a compensating planimeter. The stems and leaves were then cut into small pieces to facilitate drying and their dry weights obtained in the usual way.

## EXPERIMENTAL RESULTS

*Dry weights and leaf areas.* Since green weight yields, dry weight yields, leaf areas, stem diameters and height of stems were obtained at the termination of each of the three series, five sets of growth measurements are available for each culture. However, only two sets of these measurements (dry weights and leaf areas) will here be presented, since the dry weight yields followed by leaf areas may safely be regarded as the most reliable of the five criteria, and since the remaining three, in a general way, show the same relationships as do these two. When dry weights are taken as a standard and the correlation coefficients of the others are calculated according to the formulae as given by Yule (1922), the leaf area records show the highest correlation ratio ($1.00 \pm .00007$). The remaining records in the descending order of the value of their correlation coefficients are green weights ($1.00 \pm .0027$), stem heights ($.964 \pm .0045$), and stem diameters ($.860 \pm .016$). The numerical data of dry weight yields and of leaf areas for the plants of each of the three series are presented in table 3. The data in the columns marked 'average' are the values obtained by averaging corresponding data from the three series, each average value thereby involving three cultures and eight plants. Each value in the table is relative to the corresponding value of culture 1, taken as 100. The actual yield values for this culture are given in parentheses, in grams for dry weights, and in square centimeters for leaf areas. The actual yield measurements for any culture may be obtained by multiplying its relative value by the corresponding actual value given in parentheses for culture 1.

The response of the rhododendron plants to the different salt proportions of various solutions to which they were exposed in the sand cultures, will here be considered with reference to the range in the proportions of each salt within which high average quantitative growth measurements for this species were obtained. These ranges for high yields will be compared with the corresponding ranges for the entire series.

From the data of table 3, it will be observed that a fairly wide range of relative yield values of dry weights and leaf areas occurs within each series, which may be in a large measure, if not entirely, attributed to the variations in the relative salt proportions from culture to culture in the respective series. Since low and medium values hold little of interest or value in this connection, they will not be considered here.

The highest five average measurements of dry weight yields and leaf areas, and the salt proportions of the solutions which produced these, together with the ranges from the highest to the lowest of each set of values, have been brought together in table 4. For facility in making comparisons, the table also shows the maximum, minimum, and range values

of the partial volume-molecular salt concentrations and growth measurements for the entire series.

It is to be pointed out that the highest five yields in the three different series were not always produced by corresponding cultures, as is indicated by the data of table 3. Only one culture within the range of those pro-

TABLE 3

*The relative values of the dry weights of tops and of leaf areas of plants grown in four-salt solutions, all having a total osmotic concentration of 1.0 atmosphere, but differing from each other in the proportion of the four salts*

| CULTURE NUMBER | RELATIVE GROWTH VALUES (AVERAGE PER PLANT) | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | DRY WEIGHT OF TOPS | | | | LEAF AREAS | | | |
| | SERIES A | SERIES B | SERIES C | AVERAGE | SERIES A | SERIES B | SERIES C | AVERAGE |
| 1 | 1.00 (5.49) | 1.00 (26.8) | 1.00 (15.7) | 1.00 (16.0) | 1.00 (685) | 1.00 (1820) | 1.00 (1264) | 1.00 (1256) |
| 2 | 0.73 | 1.26 | 0.73 | 0.91 | 0.80 | 1.20 | 0.76 | 0.92 |
| 3 | 0.76 | 1.66 | 0.63 | 1.02 | 0.74 | 1.60 | 0.63 | 0.99 |
| 4 | 0.72[a] | 1.26 | 0.65 | 0.88 | 0.82 | 1.17 | 0.66 | 0.88 |
| 5 | 1.07 | 1.63 | 1.38 | 1.36 | 1.10 | 1.37 | 1.26 | 1.24 |
| 6 | 0.82 | 1.66 | 1.22 | 1.23 | 0.78 | 1.37 | 1.12 | 1.09 |
| 7 | 1.09 | 1.08 | 0.59[a] | 0.92 | 1.02 | 1.09 | 0.53 | 1.05 |
| 8 | 0.94 | 1.10 | 0.84 | 0.96 | 0.97 | 1.13 | 0.91 | 0.99 |
| 9 | 1.31[a] | 1.82 | 1.13[a] | 1.42 | 1.10[a] | 1.62 | 1.08[a] | 1.27 |
| 10 | 0.85 | 1.35 | 1.01 | 1.07 | 0.97 | 1.37 | 1.09 | 1.14 |
| 11 | 1.18[a] | 0.84 | 1.32 | 1.11 | 1.22[a] | 0.92 | 1.25 | 1.13 |
| 12 | 0.90 | 1.82 | 1.00 | 1.24 | 0.78 | 1.56 | 0.98 | 1.11 |
| 13 | 0.63 | 1.47 | 0.94 | 1.01 | 0.67 | 1.34 | 0.86 | 0.96 |
| 14 | 0.93 | 1.51 | 1.40 | 1.28 | 0.96 | 1.54 | 1.41 | 1.30 |
| 15 | 0.88 | 1.71 | 0.91 | 1.17 | 0.86 | 1.57 | 0.89 | 1.11 |
| 16 | 0.83 | 1.45 | 1.02 | 1.10 | 0.87 | 1.44 | 1.06 | 1.12 |
| 17 | 1.07 | 1.39 | 1.04 | 1.17 | 0.96 | 1.50 | 1.10 | 1.19 |
| 18 | 0.77 | 1.43 | 0.94 | 1.05 | 0.76 | 1.38 | 0.86 | 1.00 |
| 19 | 0.88 | 1.70 | 0.96 | 1.18 | 0.81 | 1.65 | 1.02 | 1.19 |
| 20 | 0.96 | 1.47 | 1.03 | 1.15 | 1.05 | 1.41 | 1.04 | 1.17 |

[a] Two plants per culture.

ducing high yields is common to the three series. This is culture 9. This lack of agreement between the cultures of the three series may be accounted for by the fact that only one series was conducted during any one experimental period. The three series were carried out during three different years and were not begun nor terminated at the same season of the year. Of course, it was not within the province of this investigation to evaluate the influence of seasonal change or environmental differences upon growth as related to the inorganic nutrition of the rhododendron.

Considering the average dry weight criterion, it will be observed from table 4 that the volume-molecular proportions of the four salts characterizing the culture solutions which produced high dry weight yields, without exception, are limited to certain ranges which are less extensive than the corresponding total ranges for the entire series. The range in the propor-

tions of $KH_2PO_4$ and $Ca(NO_3)_2$ which produced high yields are relatively narrow in comparison with the corresponding ranges for the entire series. The proportions of $(NH_4)_2SO_4$ and $MgSO_4$ show a relatively wide range within which high dry weight yields were produced.

The range in average dry weight values for the five cultures producing high yields is quite narrow, extending from 22.5 grams to 24.6 grams per

TABLE 4

*Volume-molecular salt proportions and ranges of these for the solutions which produced the highest five average measurements of dry weight and leaf areas*

| | CULTURE NUMBERS | PARTIAL VOLUME-MOLECULAR CONCENTRATIONS | | | | AVERAGE GROWTH MEASUREMENTS PER PLANT | |
|---|---|---|---|---|---|---|---|
| | | $KH_2PO_4$ | $Ca(NO_3)_2$ | $MgSO_4$ | $(NH_4)_2SO_4$ | DRY WEIGHT GMS. | LEAF AREAS SQ. CM. |
| Cultures Producing Highest Five Dry Weight Yields | 5 | .00211 | .00146 | .01185 | .0042 | 23.4 | |
| | 6 | .00211 | .00438 | .00711 | .0042 | 22.7 | |
| | 9 | .00211 | .00438 | .00237 | .0070 | 24.6 | |
| | 12 | .00633 | .00438 | .00711 | .0014 | 23.1 | |
| | 14 | .00633 | .00146 | .00711 | .0042 | 22.5 | |
| | Range | .00422 | .00292 | .00948 | .0056 | 2.1 | |
| Cultures Producing Highest Five Yields of Leaf Area | 5 | .00211 | .00146 | .01185 | .0042 | | 1612 |
| | 9 | .00211 | .00438 | .00237 | .0070 | | 1690 |
| | 14 | .00633 | .00146 | .00711 | .0042 | | 1744 |
| | 17 | .01055 | .00146 | .00711 | .0014 | | 1592 |
| | 19 | .01055 | .00146 | .00237 | .0042 | | 1615 |
| | Range | .00844 | .00292 | .00948 | .0056 | | 152 |
| Entire series | Maximum | .01477 | .01022 | .01659 | .0098 | 24.6 | 1744 |
| | Minimum | .00211 | .00146 | .00237 | .0014 | 14.7 | 1118 |
| | Range | .01266 | .00876 | .01422 | .0084 | 9.9 | 626 |

plant, with a range of 2.1. This indicates that the five culture solutions which produced the highest five average yields of dry plant material show nearly equal efficiency in producing good rhododendron plants under the experimental conditions here described.

Turning now to the criterion of average leaf areas, the data of table 4 show that the ranges in the molecular proportions of $Ca(NO_3)_2$, $MgSO_4$ and $(NH_4)_2SO_4$ in the solutions which produced the highest five average yields of leaf areas are identical with those of the solutions which produced the highest five average dry weight yields. While the molecular proportions of $KH_2PO_4$ in the cultures which produced high average yields of dry plant material show a relatively narrow range, those of the solutions corresponding to the highest five average leaf area measurements show a relatively wide range as compared with the total range for the entire series.

The range in the absolute average leaf areas for the cultures yielding the highest five measurements is quite narrow and bears approximately the same relation to the total range for the entire series as does the range in dry weights, for the five high yielding cultures, to the corresponding range for the entire series. Three cultures in the group of five which gave the highest average leaf area measurements are also in the group of five cultures which produced high average dry weight yields. These cultures are 5, 9, and 14, corresponding to the solution numbers in table 1.

Fig. 1. Plants in two cultures selected from the best five (left), and two selected from the poorest five (right) in series B at the termination of the experiment. Age of plants 16 months from seed.

It is to be emphasized that while the highest five average yields produced by the solutions of the series employed were much superior to the lowest five average yields, not a single set of salt proportions in the entire series failed to produce what might be regarded as fairly good plants. This is clearly indicated in figure 1, which shows photographs of two cultures selected from the best five in Series B and two selected from the poorest five, at the termination of the series. No pronounced injurious effects which could be traced directly to an unbalanced condition of salt proportions, appeared in any culture of the series. While the plants of some cultures were much smaller than those of others, as the data indicate, and were considerably retarded in development as compared with the plants which made optimum growth in the series, they were, nevertheless, healthy and vigorous plants. This is an indication that within certain limits *Rhododendron ponticum* is not particularly sensitive to variations in

salt proportions, so long as the extreme variations do not actually involve an absolute deficiency of an essential element.

From the data presented in table 4, it appears that the poor growth of rhododendron so often encountered in the field is probably not due to an unfavorable balance of the elements required in major quantities for growth, since the species is tolerant of a fairly wide range in the proportions of these elements. This evidence tends to indicate that there are other factors which are operative here. One of these factors, however, may still be nutritional in nature, as the retarded growth may be due to a deficiency of some one or more of the essential ions required in minor quantities for growth.

*Apparent condition of plants.* It is a matter of some interest to note, as already pointed out, that no combination of the salts employed produced what might be termed a severe pathological condition in the plants. During the course of the three series, however, five plants out of a total of 160 died, but this was due to originally diseased stock and not to any severe nutritional disturbance, inasmuch as the remaining plants in each of the cultures containing the originally injured one survived and made excellent growth.

While none of the experimental treatments resulted in pronounced injury, it was obvious that for the rhododendron some of the solutions were poorly balanced with respect to the proportions of the four constituent salts. The only obvious injury which appeared manifested itself in a chlorotic condition of the plants in several cultures. This chlorosis appeared at an early stage in the plants of corresponding cultures in each of the three series. These cultures were 3, 4, 7, and 13, each having relatively high proportions of calcium nitrate and low proportions of the three other salts. The chlorotic condition of the plants was readily traced to their inability to obtain and utilize efficiently the iron from the solutions supplied to these cultures. By maintaining relatively high concentrations of iron in the solutions, the chlorotic condition of the plants in three of the cultures completely disappeared. The plants in culture 4, however, in which the solution contained the maximum proportion of calcium nitrate and the minimum proportions of each of the other three salts, remained chlorotic throughout the growth period.

It is of interest to point out that although the solutions were continuously renewed, the roots of the slow-growing rhododendron produced considerable change in the reaction of the medium during the absorbing process. In table 5 are recorded the hydrogen-ion concentrations, expressed as pH, of the twenty culture solutions of Series B at different stages in the growth period after the roots of the plants had been in con-

tact with the solutions in the sand cultures for a definite interval of time.

In preparation for making pH determinations, each sand culture was flooded with distilled water and then allowed to drain until dripping from the hole in the bottom of the container ceased. This was done to remove as much as possible of the old solution from the sand and roots of the plants. New solution was then percolated through the sand for a period of 24 hours by the continuous flow method at the rate of one liter in 24 hours.

TABLE 5

*Hydrogen-ion concentrations, expressed as pH, of the culture solutions and leachings from the cultures at various stages of growth*

| CULTURE NUMBER | HYDROGEN-ION CONCENTRATION, EXPRESSED AS pH | | | |
|---|---|---|---|---|
| | CULTURE SOLUTION (ORIGINAL) | LEACHINGS FROM CULTURES AFTER VARIOUS INTERVALS OF GROWTH | | |
| | | 9 WEEKS | 16 WEEKS | 21 WEEKS |
| 1 | 4.46 | 3.80 | 3.46 | 3.65 |
| 2 | 4.48 | 3.90 | 3.65 | 3.58 |
| 3 | 4.47 | 3.82 | 3.71 | 3.51 |
| 4 | 4.53 | 3.83 | 3.73 | 3.46 |
| 5 | 4.56 | 3.82 | 3.45 | 3.45 |
| 6 | 4.53 | 3.84 | 3.61 | 3.57 |
| 7 | 4.55 | 3.87 | 3.62 | 3.67 |
| 8 | 4.53 | 3.93 | 3.53 | 3.66 |
| 9 | 4.48 | 3.89 | 3.66 | 3.49 |
| 10 | 4.55 | 4.00 | 3.57 | 3.56 |
| 11 | 4.35 | 3.86 | 3.48 | 3.63 |
| 12 | 4.30 | 3.97 | 3.55 | 3.67 |
| 13 | 4.29 | 4.11 | 3.45 | 3.46 |
| 14 | 4.29 | 3.83 | 3.47 | 3.84 |
| 15 | 4.30 | 4.02 | 3.63 | 3.59 |
| 16 | 4.31 | 3.88 | 3.52 | 3.47 |
| 17 | 4.18 | 3.89 | 3.51 | 3.59 |
| 18 | 4.19 | 3.97 | 3.82 | 3.82 |
| 19 | 4.23 | 3.77 | 3.36 | 3.69 |
| 20 | 4.15 | 3.69 | 3.53 | 3.60 |

Samples of the drip from the opening in the bottom of each pot were then collected and the pH of the solutions determined electrometrically.

It will be observed that the solutions of the different cultures in the series are all rendered quite acid through differential absorption of essential ions, and indeed this condition appears to be necessary, under the conditions of these experiments, to avoid chlorosis in the plants due to an apparent deficiency of available iron. However, the rhododendron apparently absorbs and assimilates iron only with some difficulty, particularly from a medium the pH of which approaches the point of neutrality. It is here suggested that this may be a direct effect of relatively high pH values of the medium upon the absorbing and assimilating power of the plant, which may render the absorption and assimilation of iron by the rhododendron quite difficult, and may not be entirely the result of iron de-

ficiency due to precipitation from solution. This is given as a suggestion and not as a fact established by experimental evidence.

By special methods it has been possible to produce excellent rhododendron plants by the use of balanced solutions of the series here employed, at pH values considerably above the point of complete precipitation of inorganic iron, as established by the work of Patten and Mains (1920). This was accomplished as follows: Solution $T_8R_1C_1$ of the series was used as the culture medium. The pH of this solution was adjusted to 6.5–7.0 by the partial substitution of the di-basic potassium phosphate for the monobasic form. With this solution so adjusted, the plants were grown in sand cultures in precisely the same manner as were the plants of the regular series, except that the rate of continuous flow of the solution was somewhat higher, in order to avoid any pronounced reduction in pH of the solution through contact with the plant roots. Twice a week each of the cultures was flooded with one liter of distilled water and allowed to drain, to remove most of the solution from the sand and roots of the plants. 400 cc. of an aqueous solution of $FeSO_4$ (100 p.p.m. of iron) was then poured upon the sand in the culture. This was allowed to remain thirty minutes, after which the continuous flow method of renewal of culture solution was resumed.

Fig. 2. Plant on the right grown in soil; age two years, height 0.76 meters. Plants on the left grown in sand culture; age two years, height 2.4 meters.

With this modification in the cultural method, excellent rhododendron plants were grown without the appearance of chlorosis. Without this modification in the cultural method, the use of solutions at pH 6.0–7.0 resulted only in severely chlorotic plants and ultimately in the death of the plants. Thus, in the absence of the culture solution these plants were able to absorb, from a dilute aqueous solution of iron, and assimilate during the brief interval of thirty minutes, sufficient of this element to satisfy their requirements for normal growth and development. In the presence of the culture solution at pH 6.0–7.0 and with an adequate concentration of iron in the medium, the plants failed utterly to absorb or to assimilate the necessary supply of this element for normal growth and development.

*Observations on older rhododendron plants grown in sand cultures.* In the time period during which the three experimental series above de-

scribed were conducted, certain observations were made on older rhododendron plants which had been grown continuously in sand cultures by the method here described. Seedlings from seed germinated in sand in February, 1929 were transferred in November of the same year to the sand in three-gallon percolators, like those employed in the regular series. Here they were grown in the greenhouse in sand continuously supplied with solution $T_5R_1C_1$ by the continuous flow method until they bloomed in July 1931 at the age of two and one-half years. These plants again bloomed in February 1932, at the age of three years. By a successive development of the individual florets in the clusters, this second bloom continued over a period of seven weeks.

In figure 2 are shown photographs of two of these plants at the age of nearly two years, compared with one of the same age grown in a good soil—(sand-leaf-mold-mixture) side by side with the two grown in sand cultures. These three plants were transplanted as seedlings at the age of five months, from the same seed bed of sand at the same time. The plants grown in sand cultures continuously supplied with a culture solution averaged 2.4 meters in height; those grown in the soil-sand-leaf-mold-mixture showed an average height of 0.76 meters.

In May 1931, six plants from this same group which had been grown by the sand culture method, were removed from the sand cultures in which they had been growing, the surplus sand was shaken from their roots, and in this condition they were transplanted out-of-doors in ordinary garden soil. When transplanted into the open they averaged nearly one meter in height. Each of these six plants not only survived the transplanting in excellent condition, but also made very good growth during the summer and fall months, in spite of, or perhaps in a stricter sense, because of the rapid forcing by means of the sand culture treatment which they had received during the first 15 months of their growth. With slight protection all of these plants survived the winter out-of-doors in excellent condition, although this species is not considered entirely hardy in this latitude. Some of these plants developed excellent flower buds and bloomed during the season of 1932, at the age of less than two and one-half years from seed, and produced mature seeds in the fall of 1932.

The ability of these plants to withstand the shock of transplanting and to endure the rigors of the hot summer months in an exposed position in the open, without serious check to growth and development, may be attributed to the remarkable root systems which the plants produce under the influence of the sand culture method with continuous solution flow. The plants under these growing conditions produce very elaborate systems of vigorous, finely-branched, fibrous roots, which expose to the

growth medium unusually extensive absorbing surfaces. These roots provide most efficient absorbing systems, and their efficiency is abundantly reflected in the vigor of top growth, as may be observed from figure 3, which shows a photograph of an average plant at the age of two years and eight months.

During the entire period of development, the seedlings employed in these experiments, grown from seeds germinated in sand, remained free from the common diseases of rhododendrons, although no special precautions were taken to prevent disease of any kind.

Fig. 3. Average plant grown in sand culture with continuous solution flow. Age two years eight months from seed.

It is thus clear that the sand culture method of seed germination and early growth of the seedlings of this species offers not only certain advantages in the development of experimental plants for purposes of investigation, but also advantages which may be usefully applied in commercial production.

SUMMARY

Seedlings of *Rhododendron ponticum* were grown in sand cultures continuously supplied with culture solutions by a method of continuous solution renewal at constant rates. Twenty different sets of salt proportions of the four component salts $KH_2PO_4$, $Ca(NO_3)_2$, $MgSO_4$, and $(NH_4)_2SO_4$ were simultaneously tested, all at a total osmotic concentration of one atmosphere. The results may be briefly summarized as follows:—

1. Volume-molecular proportions of the four salts characterizing the five solutions which produced the plants yielding the highest five average measurements of dry weight in a series of twenty cultures, are limited to certain ranges which are considerably less extensive than the corresponding total ranges for the entire series.

2. For these five solutions, ranges in the proportions of $KH_2PO_4$ and $Ca(NO_3)_2$ are relatively narrow, while those of $MgSO_4$ and $(NH_4)_2SO_4$ are relatively wide in comparison with the corresponding ranges for the entire series.

3. These five solutions show nearly equal efficiency in producing good rhododendron plants.

4. Within certain limits, *Rhododendron ponticum* is not particularly sensitive to variations in salt proportions, so long as these do not actually involve an absolute deficiency of one of the essential elements. Not a single set of salt proportions in a series of twenty, produced what might be regarded as a severe pathological condition in the plants.

5. The sand culture method of seed germination and early growth of seedlings of *Rhododendron ponticum* offers advantages which may be usefully applied in the development of experimental plants and in commercial production.

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# A revision of the section Gymnocaulis of the genus Orobanche

DAISY M. ACHEY

(WITH FIFTEEN TEXT FIGURES)

This revision has been done under the direction of Dr. Philip A. Munz of Pomona College, to whom I hereby acknowledge my deep indebtedness. I wish also to express my appreciation to those in charge of the following herbaria from which material was borrowed: Gray Herbarium of Harvard University (G), University of California (C), Missouri Botanical Garden (M), United States National Herbarium (U. S.), Pomona College Herbarium (P). The abbreviation given with each name above is used in citing specimens. My gratitude is also due to Mr. W. N. Suksdorf of Bingen, Washington for his kindness in lending type material of the species he has described.

As here treated the section Gymnocaulis includes the same plants as in Beck's recent revision of the genus *Orobanche* (Das Pflanzenreich $4^{261}$: 46. 1930). It consists entirely of North American plants. But after careful study of much herbarium material and after making dozens of camera lucida drawings of the flowers, I would treat somewhat differently the taxonomy of the plants in this section and would recognize but two species, *O. uniflora* and *O. fasciculata*, with their several varieties. The diagnostic characters for these species are quite definite and the two are easily separated from each other. But an attempt at the classification of the species into varieties is not so satisfactorv. In any given character studied there is a gradation from one extreme to the other. For example, there is a continuous series of plants from those with glabrous to those with densely pubescent anthers. The color, size, and shape of the floral parts were found to be as variable as the pubescence on the anthers. However, the differences in the plants of a given species were too marked to make it seem advisable to place them all together. I have separated them, therefore, on the basis of the most constant characters observed, while at the same time recognizing the high degree of intergradation between the varieties.

The divisions were more definite in the *uniflora* than in the *fasciculata* group. In *O. uniflora*, I found considerable intergradation between the three western varieties: *purpurea*, *Sedi*, and *minuta*. But these three separated nicely, both morphologically and geographically, from the two eastern varieties. Suksdorf (Allg. Bot. Zeitsch. **12**: 27. 1906) would recognize *Aphyllon inundatum* as a fourth western entity. But there seems to me to be more variation within the variety *Sedi* than between the types of *Sedi*

and *inundatum*, therefore I have put them together. The fact also that their geographical distribution is practically the same adds to my feeling that the two are not different varieties.

My greatest difficulty has come in trying to divide the *fasciculata* group into varieties. I was hesitant about throwing them all together, for, considered in their extremes, the plants were decidedly different. For example, some were dark with small corollas and glabrous anthers, while others were light with large corollas and pubescent anthers. But these combinations of characters were not constant enough to warrant being used as criteria in making varieties. Even in the same collection, one might find any variation of these two extremes. Therefore, after making several unsuccessful attempts at getting any clear-cut distinctions within the group, it seemed advisable to divide them into three varieties which have forms intergrading with one another. In some cases I might have listed more intermediate forms than I have done. However, I think that I have cited enough to give one an idea of the high degree of variability between the three varieties.

### GYMNOCAULIS

As section, Nutt. Gen. N. Am. **2**: 59. 1818; G. Beck, Pflanzenreich **4**[261]: 46. 1930. *Aphyllon*, as genus, Mitchell, Act. Phys. Med. Acad. Nat. Cur. **8**: 221. 1748; as sect., Beck, Mon. Orob. 74. 1890. *Thalesia*, as genus, Raf. Am. Mo. Mag. 267. 1818. *Loxanthes*, as genus, Raf. Neogenyt. 3. 1825. *Anoplon*, as genus, Reichb. Consp. 212b. 1828. *Anoplanthus*, sect. *Euanoplon*, Endl. Gen. Pl. 727. 1836–40.

Flowers long-pediceled, erect, from axil of bract; calyx ebracteolate, 5-lobed, campanulate, almost regular; corolla slightly bilabiate, the lobes subequal; stigma peltate; capsule 1-celled, 2-valved, with 4 placentae.

#### KEY TO SPECIES

Pedicels 1–3, two or three times length of stem; calyx lobes longer than tube..... 1. *O. uniflora*

Pedicels many, equal to or shorter than stem; calyx lobes equal to or shorter than tube. 2. *O. fasciculata*

1. OROBANCHE UNIFLORA L. Sp. Pl. 633. 1753.

Stems slender, 2–4 mm. thick, ca. 3–5 cm. long; pedicels 3 to 15 cm. long; bracts 2–6, crowded on stem; corolla 15 to 30 mm. long, lobes rounded or oblong, rarely pointed; anthers glabrous to pubescent.

#### KEY TO VARIETIES

Calyx lobes scarcely longer than tube, lanceolate, tapering gradually from base to apex. Plants east of Rocky Mts.

Ciliation on corolla lobes ca. 0.75 mm. long. On mainland. 1a. *O. uniflora var. typica*

Ciliation on corolla lobes ca. 0.5 mm. long. In Newfoundland..... 1b. *O. uniflora var. terrae-novae*

Calyx lobes about twice as long as tube, narrowly subulate from a broad base. Plants from Rocky Mts. and West.

Anthers usually pubescent; corolla 22–30 mm. long, tube 5–8 mm. wide at the throat..... 1c. *O. uniflora var. purpurea*

Anthers glabrous; corolla usually 15–22 mm. long, tube 3–5 mm. wide at the throat.

Corolla purple; parasitic on Saxifragaceae..... 1d. *O. uniflora var. minuta*

Corolla yellow or tinted with lavender; parasitic on *Sedum* or Compositae.. 1e. *O. uniflora var. Sedi*

1a. **O. uniflora** L. var. **typica,** n. nom.

*O. uniflora* L., l.c.; G. Beck, Mon. Orob. 74: 1890, in part; *Anoplanthus uniflorus* Endlicher, Gen. Pl. 727. 1836–40; *Aphyllon uniflorum* Gray, Man. Bot. ed. 1, 290. 1848; Syn. Fl. $2^1$: 312. 1878; *Thalesia uniflora* Britton, Mem. Torrey Club **5**: 298. 1894; *O. biflora* Nutt. Gen. N. Am. **2**: 59. 1818; *Phelipaea biflora* Sprengel, Syst. **2**: 818. 1825; *Anoplon biflorum* G. Don, Syst. $4^4$: 634. 1838.

Pedicels usually paired, 6–15 cm. long, 0.5 to 0.75 mm. thick; corolla 2–3 cm. long, yellowish, often with lavender lines in the throat and azure on the margins; the lobes obovate, 3–5 mm. long, strongly ciliated on margins; throat scarcely enlarged, tube slightly constricted in upper part; anthers only slightly pubescent.

Type locality, "Habitat in Virginia." Ranging throughout the Atlantic States and west to the Rocky Mt. States. Representative material, MACKINAW ISLAND: Arch Rock, *Wright* in 1915 (G). ONTARIO: Agawa River, *Pease 18957* (G). MAINE: Orono, *Fernald 2438* (G); Westbrook, *Ricker 54* (U. S.). NEW HAMPSHIRE: Groton, *Steele* in 1882 (P). VERMONT: Rutland, *Eggleston 2589* (M). MASSACHUSETTS: Cummington, *Williams* in 1907 (G); Cambridge, *Tuckerman 814* (U. S.). CONNECTICUT: Norwich, *Linnsden* in 1884 (C). NEW YORK: Peaked Mts., Washington Co., *Ann* in 1900 (P); Ithaca, *Munz 366* (P); Brooklyn, *Hicks* in 1886 (G). RHODE ISLAND: Johnstown, *Collins* in 1890 (G, U. S.). NEW JERSEY: Milltown, *MacKenzie 3048* (M). PENNSYLVANIA: Lancaster, *Small* in 1890 (M, U. S.); Sunneyton, *Smith* in 1905 (C). MARYLAND: Baltimore, *Steitz* in 1894 (U. S.); College Park, *Pond* in 1901 (G). DISTRICT OF COLUMBIA: WASHINGTON, *Steele* in 1897 (G); Seven Locks, *Steele* in 1898 (U. S.). VIRGINIA: Charlestown, *Pringle* in 1876 (P). WEST VIRGINIA: Upshur, *Nutter* in 1897 (M, U. S.). NORTH CAROLINA: Asheville, *McCarthy* in 1888 (U. S.). SOUTH CAROLINA: Keowee, *House 1896* (U. S.). FLORIDA: Jacksonville, *Smith 445* (U. S.). MISSISSIPPI: Tchula, *Woodsen & Anderson 1519* (M). KENTUCKY: Natural Bridge, *McFarland 76* (M). TENNESSEE: Knoxville, *Ruth* in 1898 (M, U. S.). OHIO: Brady Lake, Portage Co., *Webb* in 1923 (G). ILLINOIS: Park Ridge, *Walker* in 1905 (U. S.). MICHIGAN: Whitmore Lake, Washtenaw Co., *Chandler* in 1915 (U. S.). IOWA: Ames, *Stewart* in 1893 (M). NEBRASKA: Lincoln, *Williams* in 1886 (U. S.). KANSAS: Prairie, Riley Co., *Hansen 1007* (G, M, U. S.). MISSOURI: Courtney, *Bush 7100* (C, G, M, U. S.); St. Louis, *Engelmann* in 1842 (M). OKLAHOMA: Leflore Co., *Blakley 3436* (G). ARKANSAS: Far-

mington, *Palmer 24758* (M). MONTANA: Belt River, *Williams 239* (C); Camp 13, Southern Montana, *Rose 247* (U. S.).

The two collections of var. *typica* from Montana represent an exception to the usual absence of this variety in the Rocky Mountain States.

1b. **Orobanche uniflora** L. var. **terrae-novae** (Fernald), n. comb.

*Orobanche terrae-novae* Fernald, Rhodora **28**: 235. 1926.

Very similar to var. *typica*. Peduncles about 1 mm. thick; corolla lobes oblong, 5-7 mm. long.

Type locality, Bard Harbor, St. John Bay, Newfoundland. Ranging throughout Newfoundland. Material seen—NEWFOUNDLAND: Bard Harbor, *Wiegand et al. 29049*, type specimen, (G); Stanleyville, *Fernald et al. 2014* (G); Killdevil, *Fernald et al. 2015* (G); Hannah's Head, *Fernald & Long 2016* (G); St. John Island, *Fernald et al. 29050* (G); Burnt Cape, *Fernald & Long 29051* (G); Coal River, *Waghorne* in 1886 (G); Grand Falls, *Fernald & Wiegand 6211* (G); Birchy Cove, *Fernald & Wiegand 4018* (G); Doctor Hill, *Fernald & Long 29052* (G); Harbor Hill, *Fernald & Long 29053* (G); Penguin Hill, *Fernald et al. 2013* (G).

Fernald gives specific rank to *O. terrae-novae*. But he suggests that "it is merely a geographic variety," and my observations tend to verify this statement. The characters separating the two groups seem too insignificant to warrant giving more than varietal rank to *O. terrae-novae*. Fernald calls attention to two linear yellow lines on the throat. But Nuttall gives this (Gen. N. Am. **2**: 59. 1818) as a character of *O. biflora* (placed here as a synonym of var. *typica*). According to Fernald *O. terrae-novae* is odorless while var. *typica* is fragrant.

An example of the similarity between the two groups may be seen in the following two sheets: South Carolina, Keowee, *House 1896* (U. S.) and Newfoundland, Birchy Cove, *Fernald & Wiegand 4018* (G). In both these sheets the length of the corolla lobes ranges from 3 to 5 mm. In the Newfoundland one, as is usual in *terrae-novae*, the ciliation tends to be longer and the pedicels thicker than in *typica*. However the lobes in the Newfoundland plant are less oblong than those of Carolina, although they should be more so. Another example of *typica* with long, oblong lobes is a specimen from Arkansas: Hotsprings, *Palmer 24506* (M.)

The following specimens from Quebec were annotated as *O. terrae-novae* by Fernald: Renard River, *Victorin & Germain 27906* (G); Chicotte River, *Victorin & Germain 27908* (G); Carbeaux, *Rousseau 26652* (G). The other specimens from Quebec he leaves as *Orobanche uniflora:* Rimouski Co., *Fernald & Collins 1154* (G); River Juniter, *Victorin & Germain 25459* (G); Stanstead, *Knowlton* in 1924 (C). But they all seem to be intergrades between *terrae-novae* and *typica*. The corolla lobes tend to be obovate and the ciliation short as in *typica*. But the pedicels are thick as in *terrae-novae*. The length of the lobes ranges between that of the two varieties, i.e. from 3 to 7 mm.

1c. **Orobanche uniflora** L. var. **purpurea** (Heller), n. comb.

*Thalesia purpurea* Heller, Bull. Torrey Club 24: 313. 1897; *Orobanche porphyrantha* Beck, Pflanzenreich 4[261]: 49. 1930.

Pedicels 3 to 10 cm. long; corolla usually deep purple, constricted at base of tube and becoming 5 to 8 mm. wide at throat.

Type locality, Lewiston, near mouth of Potlach, Idaho. Ranging from British Columbia through Washington, Oregon and Idaho to southern California. Representative material—BRITISH COLUMBIA: Jardo, *Shaw 691* (U. S.). WASHINGTON: Tacoma, *Rensselaer* in 1894 (G); Blue Mts., *Horner 403* (U. S.); Yakima region of Cascades, *Brandegee 14197* (M); Cheney, *Tucker* in 1890 (G); Wenatchee Mts., *Elmer 463* (G); Bingen, *Suksdorf 2323* (C, G, M, U. S.); Ellensburg, *Whited 327* (U. S.); Wawawai, *Elmer* in June 1897 (M). OREGON: Hood River Co., *Henderson 543* (M); The Dalles, Wasco Co., *Sheldon S. 10209* (G, M, P, U. S.); Blue Mts., *Henderson 5416* (G); Pendleton, Umatilla Co., *Gale 529* (M); Mt. Emily, *Cusick 2396* (C, G, M, P, U. S.); Rock Creek, *Leiberg 76* (G, M, U. S.). CALIFORNIA: Yreka, *Smith 638* (G, U. S.); Mendocino Co., *Bolander 4677* (C, G, U. S.); Donner Lake, *Sonne* in 1886 (C); Little Chico Canyon, *Austin* in 1897 (U. S.); Hupa, *Manning 96* (C); Mts. north of San Francisco, *Rattan 54* (G); Grahams, Humboldt Co., *Blasdale* in 1896 (C); Sierra Mts., *Davidson* in 1885 (P); Musser Hill, *Gate 354* (P); Lake Tahoe, *Chestnut & Drew* in 1890 (C); Santa Cruz Mts., *Pendleton 278* (U. S.); Long Valley, *Brewer 4677* (U. S.); Tamarack Lake, *Essig* in 1928 (C). IDAHO: Coeur d'Alene, *Rust 114* (U. S.); Lewiston, Nez Perces Co., *Heller 3099*, type collection, (C, M, U. S.); Squaw Creek, Boise Co., *Macbride 813* (C, G, M, P, U. S.); Middle Valley, *Jones* in 1900 (P).

Some plants from Texas (without locality), *Lindheimer 489* (M, U. S.), have not been cited with this group for they might belong to the var. *typica*. The calyx lobes are lanceolate as in var. *typica*, but the plants are darker and less delicate than in *typica*. They also do not have the purple margin on the corolla lobes which is characteristic of var. *typica*. There is another collection from the immediate range of var. *purpurea* which resembles the ones from Texas: CALIFORNIA, Comptche, Mendocino Co., *Walker 368* (C). I am placing them all as doubtful specimens of var. *purpurea*, for they probably belong more nearly to this group than to any other variety of *O. uniflora*. Their long pedicels (15–20 cm.) and pubescent anthers would separate them from var. *Sedi* and var. *minuta*.

In his recent revision (Pflanzenreich 4[261]: 49. 1930) Beck keeps *O. porphyrantha* (*purpurea*) as a species distinct from *O. uniflora*. The results of my work in the group would lead me to place it as only a variety of *O. uniflora*. Beck has reduced *minuta* to a variety and var. *Sedi* to a form of *O. uniflora*. I do not find that *purpurea* differs more from *typica* than either *minuta* or *Sedi* does. It is true that var. *purpurea* is darker and its corolla tube tends to be broader than in var. *typica*. But in var. *minuta* the corolla lobes are less spreading and the plant is smaller and darker than in *var. typica*. In var. *Sedi* the lobes are truncate or oblong, not spreading and the plant is usually of smaller size, thus making it as distinct an entity from var. *typica* as var. *purpurea* is. Some specimens of var. *purpurea* are quite light while some of var. *typica* are tinged with considerable purple. The

breadth of the corolla lobes is more or less variable in both groups. For these reasons I am considering *purpurea*, as well as *Sedi* and *minuta*, as varieties of *O. uniflora*.

There is a high degree of intergradation between var. *Sedi* and var. *purpurea*. Their affinity is especially shown in the plants from Idaho which have a tendency to have lighter corollas and less pubescent anthers than most others in the var. *purpurea*. I have seen three sheets of the type collection; the one from the Missouri Botanical Garden has glabrous anthers as in var. *Sedi*. Another intermediate form is from Vancouver Island, Victoria, *Macoun 750* (M). The corolla is dark purple and shaped like var. *purpurea*, but the anthers are glabrous.

1d. **Orobanche uniflora** L. var. **minuta** (Suksdorf) G. Beck, Pflanzenreich $4^{261}$: 49. 1930.

*Aphyllon minutum* Suksdorf, Deutsch. Bot. Monatsschr. **18:** 155. 1900; *Thalesia minuta* Rydb. Bull. Torrey Club **36:** 693. 1909.

Pedicels 2–7 cm. long; corolla strongly curved, usually deep purple, tube 4–5 mm. wide at the throat, lobes rounded.

Type locality, Bingen, Klickitat Co., Washington. Ranging from British Columbia through Washington and Oregon to Southern California. Material seen—BRITISH COLUMBIA: Victoria, *Macoun 88173* (M); Vancouver Island, Ford Bay, *Carter* in 1918 (G); Mt. Finlayson, *Carter* in 1918 (G); Selkirk Mts., *Butters & Holway 268* (G); Trail, *Macoun 67881* (C). WASHINGTON: Pullman, *Elmer 43* (P), *Piper* in 1894 (M); Bingen, *Suksdorf 2345* (C, U. S., Suksdorf); Almota, *Piper 1809* (G, M). OREGON: The Dalles, *Lunell* in 1903 (G); Elk Rock, *Nelson 1268* (G); Selma, *Henderson 6008* (M, U. S.); Grants Pass, *Piper* in 1921 (G); Wimer, Jackson Co., *Hammond 320 A*, May 22, 1893 (M, U. S.). CALIFORNIA: Egg Lake, Modoc Co., *Baker & Nutting* in 1894 (C); Shasta Valley, *Butler 730* (C, P); Fisher's Cabin, Calaveras Co., *Hansen 246* (M, P); Mokelumne Hill, Calaveras Co., *Blaisdell*, (G); Panoche Pass, San Benito Co., *Abrams & Borthwick 7902* (P); Lafayette, *Davy 1004* (C); Rocky Bluff, *Bruce* in 1897 (P); San Juan, *Elmer 4635* (U. S.).

1e. **Orobanche uniflora** L. var. **Sedi** (Suksdorf), n. comb.

*Aphyllon Sedi* Suksdorf, Deutsch. Bot. Monatsschr. **18:** 155. 1900. *Thalesia Sedi* Rydberg, Bull. Torrey Club **40:** 485. 1913. *Orobanche Sedi* Fernald, Rhodora **28:** 236. 1926. *Orobanche uniflora* f. *Sedi* G. Beck, Pflanzenreich $4^{261}$: 48. 1930. *Aphyllon inundatum* Suksdorf, Allg. Bot. Zeitsch. **12:** 27. 1906. *Orobanche uniflora* f. *inundata* G. Beck, Pflanzenreich $4^{261}$: 49. 1930.

Pedicels 2–7 cm. long; corolla usually straw-colored, slightly curved, rarely spreading, tube 3–5 mm. wide at the throat, lobes truncate or oblong.

Type locality, Bingen, Klickitat Co., Washington. Ranging from British Columbia south through Washington, Oregon, California, Idaho, Utah, Montana, Wyoming and Colorado. Material seen—BRITISH COLUMBIA: Mt. Finlayson, *Pineo* in 1896 (C); Sproat, *Macoun* in 1890 (G); Victoria, *Fletcher* in 1885 (G). WASHINGTON: Bingen, *Suksdorf 2642* (U. S., Suksdorf); Rock Creek, *Sandberg & Leiberg 99* (U. S.); Blue Mts.,

*Horner 404* (U. S.); Chenowith, *Suksdorf 2130*, type collection, (C, G, M, U. S., Suksdorf); Wawawai, *Elmer 776* in May, 1897 (M, U. S.); Waitsburg, *Horner R. 397 B. 404* (G). OREGON: Clearwater, *Spalding*, (G); Abbots Butte, *Leiberg 4255* (U. S.). CALIFORNIA: Crystal Springs, San Mateo Co., *Baker 1927* (P); Donner Lake, *Sonne* in 1882 (C); Mt. Hamilton, *Rutter 19* (U. S.); Antelope, Amador Co., *Hansen 1872* (U. S.); Colby, *Austin 303* (P); Glen Alpine, Lake Tahoe Region, *McGregor 141* (U. S.); Emigrant Gap, *Jones* in 1882 (P); San Bernardino Co., *Parry & Lemmon 322* (M); Yucaipa, San Bernardino Co., *Feudge 543* (P). IDAHO: Henry Lake, Fremont Co., *Payson 2038* (G, M); Payette Lake, *Jones* in 1899 (P). UTAH: Wasatch Mts., *Watson 764* (U. S.); Uinta Mts., Summit Co., *Payson 5047* (M, P); Silver Lake, *Jones* in 1895 (P); City Creek Canyon, *Jones* in 1880 (P). MONTANA: Columbia Falls, *Williams* in 1895 (U. S.). WYOMING: Manmoth Hot Springs, *Mearns 678* (U. S.), *746* (U. S.). COLORADO: Denver, *Duthie & Clokey 3875* (G, M, P).

Fernald (Rhodora **28:** 236. 1926) states that "the species of Pacific North America with linear subulate or setaceous calyx-segments and purple-violet corolla is quite distinct from the eastern *O. uniflora*. The western plant is *Orobanche* (*Aphyllon*) *Sedi* (Suksd.), n. comb." *O. Sedi* as described by Suskdorf (Deutsch. Bot. Monatsschr. **18:** 155. 1900) is light colored, while var. *purpurea* as described by Heller (Bull. Torrey Club **24:** 313. 1897) is deep-purple. In var. *Sedi*, as compared with var. *purpurea*, the pedicels are about one-half as long; the width of the corolla tube is smaller; the corolla lobes tend to be obtuse or oblong rather than rounded; the anthers are glabrous instead of pubescent. I, therefore, separate into three varieties of *O. uniflora* (*purpurea*, *Sedi*, *minuta*) the plants included by Fernald in his *O. Sedi*.

2. OROBANCHE FASCICULATA Nutt. Gen. N. Am. **2:** 59. 1818.

Stems not slender, 3–10 mm. thick, 3–15 cm. long; pedicels 3–15 cm. long; bracts 5 to 12, not crowded on stem; corolla 15–30 mm. long, lobes pointed, rounded or truncate; anthers glabrous or pubescent.

KEY TO VARIETIES

Plant purplish; corolla 2–5 mm. wide at throat; anthers glabrous to pubescent..... 2a. *O. fasciculata var. typica*

Plant yellowish; corolla 3–8 mm. wide at throat; anthers usually pubescent.

Corolla densely pubescent, usually 15–20 mm. long; pedicels 1–6 cm. long; calyx lobes usually acute..... 2b. *O. fasciculata var. lutea*

Corolla not densely pubescent, 22–30 mm. long, 4–8 mm. wide at throat; pedicels 2–10 cm. long; calyx lobes usually acuminate..... 2c. *O. fasciculata var. franciscana*

2a. **Orobanche fasciculata** Nutt. var. **typica,** n. nom.

*O. fasciculata* Nutt. Gen. N. Am. **2:** 59. 1818; Beck, Pflanzenreich **4**[261]**:** 50. 1930; *Phelipaea fasciculata* Sprengel, Syst. **2:** 818. 1825; *Loxanthes fasciculatus* Raf. Neogenyt. 3. 1825; *Anoplon fasciculatus* G. Don. Syst. **3:** 633. 1838;

*Anoplanthus fasciculatus* Walpers, Repert. **3**: 480. 1844–45; *Aphyllon fasciculatum* Gray, Syn. Fl. **2**: 312. 1878. *Thalesia fasciculata* Britton, Mem. Torrey Club **5**: 298. 1894.

Pedicels 4–6 (rarely 10) in number, 3–10 cm. long; corolla usually purple, 15–22 mm. long, constricted at base of tube, lobes semiorbicular.

Type locality, Fort Mandon, North Dakota. Ranges from British Columbia and California to Michigan, Illinois, New Mexico and Arizona. Representative material—CANADA: Rosedale District, Alberta, *Moodie 1012* (G, U. S.); Sproat Lake, Vancouver Island, *Rosendahl 1940* (M, G). WASHINGTON: Tacoma, *Rensselaer* in 1894 (G); Ione, Pend Oreille Co., *Kreager 415* (G, U. S.); Blue Mts., *Horner 402* (G, U. S.). OREGON: Prairie City, *Henderson 5415* (G, M); Juniper Lake, Mono Co., *Hoffman* in 1930 (P); Lake Merced, *Brandegee* in 1907 (C); Montezuma Valley, *Jaeger 1926* (P); Cucamonga Peak, *Johnston 1556* (C, G, P); San Antonio Mt., *Johnston 1758* (P); San Jacinto Mts., Tamarack Valley, *Hall 2599* (C); San Bernardino Mts., *Parish 488* (U. S.); Cajon Pass, *Parish 19290* (C); Mt. Santiago, *Abrams 1849* (M); Catalina, *Minthorn* in 1920 (C). IDAHO: Weiser, Washington Co., *Jones* in 1899 (P); Picabo, Blaine Co., *Macbride & Payson 2991* (G, M, U. S.). UTAH: Lake Point, *Jones 2053* (P, U. S.); Salt Lake City, *Engelmann* in 1880 (M); Bear River, Summit Co., *Payson 4853* (P, M); Panguitch Lake, *Jones 6010* (C, M, P, U. S.). NEVADA: Washoe Mt., *Sonne* in 1888 (C); Charleston Mts., *Jaeger* in 1926 (P). ARIZONA: Chiricahua, *Blumer* in 1907 (G, M, U. S.). MONTANA: Wild Horse Island of Flathead Lake, *Jones* in 1908 (P); Bozeman, *Shear 5263* (U. S.). WYOMING: Pole Creek, *Nelson 1360* (G, M, U. S.); Headwaters of Mo. and Yellowstone rivers, *Hayden* in 1860 (M). COLORADO: Boulder, *Patterson 295* (C, G, M, U. S.); Salida, *Baker et al. 811* (M, P); Naturita, *Payson 363* (G, M). NEW MEXICO: Carrizo Mts. *Matthews* in 1892 (U. S.); Eagle Creek, Lincoln Co., *Wooton* in 1897 (U. S.). NEBRASKA: Council Bluffs, *Hayden 239* (M); Rush Creek, Deuel Co., *Rydberg 288* (U. S.). MICHIGAN: Manitou Island, *Engelmann* in 1840 (M). ILLINOIS: Dixon, *Vasey* in 1881 (M). INDIANA: Northwest, *Drew* in 1901 (C); Pine, *Chase 130* (G).

The variety *typica* has an unusually wide range and naturally a great variability, but I can not find characters permitting me to break it up. As a matter of fact the two varieties, *lutea* and *franciscana*, are not clear cut, as may be seen from the following intermediate forms.

The following intergrades with var. *lutea* have a tendency to be yellow like var. *lutea*. WASHINGTON: Breakmore, *Macoun & Herriot 2555* (P), *Flett* in 1898 (U. S.), *Merriam* in 1897 (U. S.). WYOMING: Dry Hills, *Leckie 798* (G); Alpine, Lincoln Co., *Payson & Armstrong 3397* (G, M, P).

As intergrades with var. *franciscana* may be cited the following specimens from California: (1) Santiago Peak, Orange Co., *Abrams 1847* (P), *1849* (M); there is a tendency for these to be light and not constricted at the base of the corolla tube, as is characteristic of var. *franciscana;* (2) San Jacinto Mts., *Spencer 1561* (P), *Hall 2154* (U. S.); these are shaped like var. *typica*, but the color and pubescence tends to be more like var. *franciscana;* (3) Laurel Hill Cemetery, San Francisco, *Eastwood 239* (G, U. S.); these are small and dark like var. *typica*, but otherwise they resemble var. *franciscana*, that is, they are not constricted at the base of the corolla tube and the lobes are not rounded.

2b. **Orobanche fasciculata** Nutt. var. **lutea** (Parry), n. comb.

*Phelipaea lutea* Parry, Am. Nat. **8**: 214. 1874; *Aphyllon fasciculatum* var. *luteum* A. Gray, Syn. Fl. **2**: 312. 1878; *Thalesia lutea* Rydberg, Bull. Torrey Club **36**: 693. 1909; *O. fasciculata* f. *lutea* G. Beck, Pflanzenreich $4^{261}$: 51. 1930.

Pedicels four to ten in number, 1–7 cm. long; corolla yellow and often tinged with lavender, slightly constricted at base, lobes usually acute; calyx lobes equal to or shorter than tube.

Type locality, Owl Creek, Wyoming. Ranging from Canada through eastern Washington and Oregon, to North Dakota, Nebraska, and Chihuahua. Representative material—ALBERTA: Banff, *McCalla 2199* (U. S.); Rosedale, *Moodie 58* (U. S.). WASHINGTON: Near Wilson Creek, Douglass Co., *Sandberg & Leiberg 298* (M, C, G, U. S.); Chelan, *Jones* in 1911 (P); Columbia River, Klickitat Co., *Suksdorf 2098* (C, G, M, U. S.), *2102* (C, G, M, U. S.); hills S.W. of Wallula, *Cotton 1051* (G, U. S.). OREGON: Grants Pass, *Prescott* in 1912 (M). IDAHO: Challis Creek, Custer Co., *Macbride & Payson 3321* (G, M, U. S.); Squaw Butte, Canyon Co., *Macbride & Nelson 146* (G, M); Big Potlatch River, Nez Perces Co., *Sandberg et al. 308* (G, P, U. S.). MONTANA: Suksdorf Gulch, Park Co., *Suksdorf 199* (P); Big Timber, Sweet Grass Co., *Eggleston 7946* (U. S.); Gardiner, *Mearns 1260* (U.S.). WYOMING: Mammoth Hot Springs, *Mearns 1357* (U. S.); Owl Creek, *Parry 202*, type collection, (G, M); Platte River, Martono Co., *Gooding 129* (M). COLORADO: Wet Mt. Valley, *Brandegee* in 797 (M); Fort Collins, *Baker 5348* (P); South Table Mt., Golden, *Knowlton 73* (U. S.). NORTH DAKOTA: Leeds, Benson Co., *Lunell* in 1916 (U. S.). SOUTH DAKOTA: Bald Hills, *Murdock 5200* (G). NEBRASKA: Neligh, *Bacon* in 1896 (G); Pine Ridge, *Williams 315* (U. S.). ARIZONA: Grand Canyon, *MacDougal 159* (U. S.); Mormon Lake, *MacDougal 78* (G). NEW MEXICO: Bear Mts., *Rusby 331* (M); Valley of Rio Grande, *C. C. Parry et al. 713* (U. S.). CHIHUAHUA: Sierra Madre, *Townsend & Barber* in 1899 (U. S.). SONORA: Puerta de St. Diego, *Hartman 579* (G).

As here understood, var. *lutea* is more inclusive than is generally treated. Originally said to be parasitic on grasses, it has been so considered, but specimens cited above were on *Eriogonum*, *Artemisia*, and other hosts.

I have mentioned under var. *typica* some intergrades with var. *lutea;* the following are more like var. *lutea*, but suggest var. *typica* in being dark and in the corolla lobes tending to be rounded: OREGON: Lost Prairie, *Cusick 2407* (G). COLORADO: Buena Vista, Chaffee Co., *Sheldon 557* (U. S.); Arboles, *Baker 610* (M). MONTANA: Spanish Basin, Madison Range, *Flodman 797* (M, U. S.). SOUTH DAKOTA: Custer, *Rydberg 936* (U. S.). ARIZONA: Grand Canyon, *Macbride & Payson 966* (G). NEW MEXICO: Barranca, Taos Co., *Heller 3585* (M, U. S.).

Not only do the following intergrades with var. *typica* tend to be dark, but the corolla is constricted at the base of the tube; COLORADO: Colorado Springs, *Knowlton 59* (U. S.). CHIHUAHUA: Chuchuichupa, *Hartman 703* (G, U. S.).

In the collection from Jackson Hole, Lincoln Co., Wyoming, *Payson 2193* (G, M, P), the plants from the Missouri and Pomona herbaria are light like var. *lutea*, but the one from the Gray herbarium is dark like var. *typica*. Since the corolla lobes are acute, I have placed them here with var. *lutea* rather than with var. *typica* as I did the other specimens from Lincoln Co.

2c. **Orobanche fasciculata** Nutt. var. **franciscana**, n. var.

Pedicels four to twelve, 4–10 cm. long; corolla straw-colored or tinged with purple, not constricted at base of tube, lobes rounded, truncate or pointed; calyx lobes usually longer than tube. (Pedicelli quatuor ad duodecim, 4–10 cm. longi; corolla straminea vel subpurpurea, non infra constricta, lobis suborbicularibus; lobis calycis tuba longioribus.)

Type, Mt. Tamalpais, Calif., *Rydberg 6230*, June 15, 1905, Pomona College Herbarium No. 146628. Ranging west of the Sierra Nevada from southern Oregon to San Diego Co., California. Material seen—CALIFORNIA: Yreka, Siskiyou Co., *Greene* in 1876 (M); Hupa, *Manning 97* (C); near Middle Creek Station, Shasta Co., *Heller 7954* (G, M, U. S.); Fort Bidwell, Modoc Co., *Manning 383* (U. S.); Big Valley, Modoc Co., *Baker & Nutting* in 1894 (C); Humboldt Co., *Chestnut & Drew* in 1888 (C); Little Chico, *Austin 1912* (U. S.); Benicia, *Blasdale* in 1892 (C); Kirker's Pass, Contra Costa Co., *Brewer 1064* (U. S.); Paradise, Butte Co., *Bruce* in 1897 (P); Mt. Osa, *Brewer 1245* (C, G, U. S.); Mt. Hood, *Heller* in 1902 (G); Sonoma, *Bioletti* in 1892 (C); Mt. Eddie, *Austin 251* (C); Mt. Tamalpais, *Jepson* in 1892 (C), *Heller 8398* (U. S.), *Bioletti* in 1892 (C), *Michener & Bioletti* in 1892 (G, U. S.); hill north of Diablo, *Brewer 1124* (C, U. S.); Mt. Diablo, *Bioletti* in 1894 (C); Rawhide Hill, Tuolumne Co., *Williamson 63* (P, U. S.); Yosemite Valley, *Fritchey 29* (M); Copper Creek, Fresno Co., *Clemens* in 1910 (P); Stanford University, *Rutter 172* (U. S.); Owl Creek, *Parry 203* (G); Vaca Mts. *Jepson* in 1892 (U. S.); Kenworthy, San Jacinto Mts., *Munz & Johnston 5481* (P); Laguna Camp, San Diego Co., *Munz 8356* (P); Santa Catalina Isl., *Pendleton 1353* (P); Box Springs, near Riverside, *Corwin & Johnston 2896* (P); Julian, *Brandegee* in 1894 (C). OREGON: Near Ashland, Jackson Co., *Applegate 431* (G, U. S.).

This variety has intergradation with var. *lutea* and var. *typica*. In the collection from California: Yosemite Valley, *"On State Survey" 27867* (C), the plant is yellow tinged with lavender as in var. *lutea*, and the corolla lobes do not spread as in var. *franciscana*. The specimen from California: Palm Springs, *Eastwood 3011* (G, U. S.) has glabrous anthers and is light yellow as in var. *lutea*. In the following collection, the corolla tends to be dark purple and strongly constricted at the throat as in var. *typica;* Bear Valley, San Bernardino Co., California, *Parish 3116* (U. S.). An intergrade with var. *typica* from Oregon: Crater Lake, *Coville 1481* (U. S.) is dark and the anthers are glabrous.

POMONA COLLEGE,
CLAREMONT, CALIFORNIA

Figs. 1–15. Fig. 1. Corolla limb of *O. uniflora* var. *typica*. Massachusetts: New Bedford, *Hervey* in 1890 (M). Fig. 2. Calyx of figure 1. Fig. 3. Corolla limb of *O. uniflora* var. *terrae-novae*. Newfoundland: Bard Harbor, *Wiegand et al. 29049* (G). Fig. 4. Corolla limb of *O. uniflora* var. *purpurea*. Washington: Bingen, *Suksdorf 2323* (M). Fig. 5. Calyx of figure 4. Fig. 6. Corolla limb of *O. uniflora* var. *minuta*. Washington: Pullman, *Elmer 43* (P). Fig. 7. Calyx of figure 6. Fig. 8. Corolla limb of *O. uniflora* var. *Sedi*. Washington: near Bingen, *Suksdorf 2642* (Suksdorf). Fig. 9. Calyx of figure 8. Fig. 10. Corolla limb of *O. fasciculata* var. *typica*. Utah: Panguitch Lake, *Jones 6010* (P). Fig. 11. Calyx of figure 10. Fig. 12. Corolla limb of *O. fasciculata* var. *lutea*. South Dakota: Black Hills National Forest, *Murdock 5200* (G). Fig. 13. Calyx of figure 12. Fig. 14. Corolla limb of *O. fasciculata* var. *franciscana*. California: Rawhide Hill, Tuolumne Co., *Williamson 63* (P). Fig. 15. Calyx of figure 14.

Figs. 1–15.

## INDEX TO AMERICAN BOTANICAL LITERATURE
### 1930–1933

The aim of this Index is to include all current botanical literature written by Americans, published in America, or based upon American material; the word America being used in the broadest sense.

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**Bold, H. C.** The life history and cytology of *Protosiphon botryoides*. Bull. Torrey Club **60**: 241–299. *pl. 10–19+f. 1–7*. 1 Ap 1933.

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